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INTERNATIONAL PHYSIOLOGY

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An Analytical Study Showing Association of Packed Cell Volume with the Severity of Anaemia in Adult Females

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Abstract

Aims: To correlate packed cell volume with severity of anaemia in adult females. **Setting and Design:** A hospital based prospective observational cross sectional 1000 consecutive cases of adult females to evaluate prevalence of spectrum of anaemia who fulfilled the inclusion criteria. Considering a confidence level of 95% and confidence interval of 3.1 the number of patients in our study to achieve statistical significance is 999. This was calculated by Survey System (<http://www.surveysystem.com/sscalc.htm#one>). **Statistical Analysis Used:** Considering a confidence level of 95% and confidence interval of 3.1 the number of patients in our study to achieve statistical significance is 999. This was calculated by Survey System (<http://www.surveysystem.com/sscalc.htm#one>). **Results:** Majority of the females (42.44%) were in the age group of 26–35 years followed by 31.9% in the age group of 18–25 years, 19.2% in the age group of 36–45 years, 4.9% in the age group of 46–55 years and 1.6% in the age group of 56–65 year. 410 (41%) females had mild anaemia while 440 (44%) and 150 (15%) females had moderate and severe anaemia. The mean packed cell volume (PCV) of females with mild anaemia was $29.13 \pm 2.14\%$ while the mean PCV of females with moderate and severe anaemia was $24.57 \pm 1.86\%$ and $20.35 \pm 1.12\%$ respectively. There was significant decrease in packed cell volume (PCV) with increase in severity of anaemia as per ANOVA test ($p < 0.05$). **Conclusion:** PCV (packed cell volume) and haemoglobin are considered as haematological indicators for classifying the severity of anaemia. Iron deficiency anemia is increasing in females, specially in reproductive age group of developing countries. The haematological parameters can aid in early recognition of type and cause of anaemia and thereby improve the outcome.

Keywords: Anaemia; Packed cell volume (PCV).

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Introduction

Anaemia is a major public health problem. Anaemia, is not a diagnosis in itself but is an objective for presence of disease. It is especially prevalent in women of reproductive age, particularly during pregnancy. Poor nutritional status during pregnancy is associated with inadequate weight gain, anemia, retarded fetal growth low birth weight, still births, preterm delivery, intrauterine growth retardation, morbidity and mortality rates.¹

Anemia is defined as a “ decrease in whole body red cell mass, a definition that precludes relative decreases in red blood cell count, haemoglobin, or haemocrit, which occur when the plasma volume”. In practice, the measurement of red cell mass is not easy, and anemia is usually diagnosed based on a reduction in the hematocrit (the ratio of packed red cells to total blood volume) and the hemoglobin concentration of the blood to levels that are below the normal range.²

A classification of anemia is based on underlying mechanism and according to alterations in red

cell morphology. Morphologic characteristics provide etiology clues such as red cell size (normocytic, microcytic, macrocytic), degree of hemoglobinization, reflected in the color of red cells (normochromic or hypochromic), and shape. In general, microcytic hypochromic anemias are caused by disorders of hemoglobin synthesis (most often iron deficiency) while macrocytic anemias often from abnormalities that impair the maturation of erythroid precursors in the bone marrow. Normochromic, normocytic anemias have diverse etiologies; in some of these anemias, specific abnormalities of red cell shape provide an important clue about cause.

Materials and Methods

A hospital based cross sectional study was done at our tertiary care centre in central clinical laboratory of tertiary care Centre, Pune among 1000 adult females to evaluate prevalence of spectrum of anaemia.

Study Design: A hospital based prospective observational cross sectional study

Study Population: 1000 consecutive cases of adult females to evaluate prevalence of spectrum of anaemia who fulfilled the inclusion criteria.

Sample Size: 1000 patients

Considering a confidence level of 95% and confidence interval of 3.1 the number of patients in our study to achieve statistical significance is 999. This was calculated by Survey System (<http://www.surveysystem.com/sscalc.htm#one>). The Survey System ignores the population size when it is "large" or unknown. Population size is only likely to be a factor when you work with a relatively small and known group of people (e.g., the members of an association). Hence a sample size of 1000 was considered adequate for our study.

Criteria of Anaemias

- Adult Non Pregnant Females HB Less Than 12 gm%.
- Adult Pregnant Females HB Less Than 11 gm%

Inclusion Criteria

- Adult females Age 18 yrs or above.
- Anaemia in non pregnant and pregnant adolescent girls.

Exclusion Criteria

- Adult females who are on treatment of anaemia.
- Females less than 18 yrs.

Methodology

Institutional ethical committee (IEC) clearance was obtained before start of study. The study was carried at ccl of tertiar care centre, Pune for a period of 2.5 yrs 2017–2019. Total of 1000 cases of adult females that came for ccl were studied. The sample for test were collected in edta tube. The slides were prepared and smears made.

Results

A hospital based cross sectional study was done among 1000 adult females to evaluate prevalence of spectrum of anaemia.

Distribution of Females According to Age

Majority of the females (42.44%) were in the age group of 26–35 years followed by 31.9% in the age group of 18–25 years, 19.2% in the age group of 36–45 years, 4.9% in the age group of 46–55 years and 1.6% in the age group of 56–65 year [Table 1] [Fig. 1].

Table 1: Distribution of females according to Age

Age (years)	N	%
18–25 years	319	31.9%
26–35 years	424	42.4%
36–45 years	192	19.2%
46–55 years	49	4.9%
>55 years	16	1.6%
Total	1000	100%

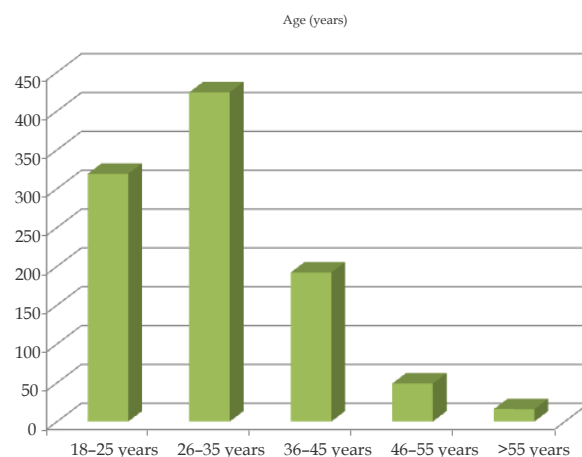


Fig. 1: Distribution of females according to age.

Distribution of Females According to Severity of Anaemia

410 (41%) females had mild anaemia while 440 (44%) and 150 (15%) females had moderate and severe anaemia [Table 2] [Fig. 2].

Table 2: Distribution of females according to severity of anaemia

Severity of Anaemia	N	%
Mild	410	41%
Moderate	440	44%
Severe	150	15%
Total	1000	100%

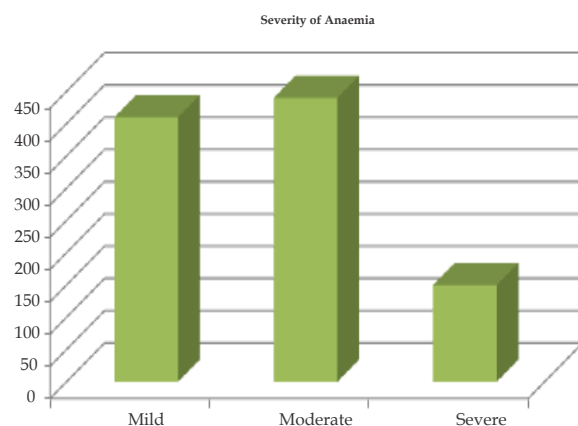


Fig. 2: Distribution of females according to Severity of Anaemia.

Association of Packed Cell Volume (PCV) and Severity of Anaemia of Females

The mean packed cell volume (PCV) of females with mild anaemia was $29.13 \pm 2.14\%$ while the mean PCV of females with moderate and severe anaemia was $24.57 \pm 1.86\%$ and $20.35 \pm 1.12\%$ respectively. There was significant decrease in packed cell volume (PCV) with increase in severity of anaemia as per ANOVA test ($p < 0.05$) [Table 3] [Fig. 3].

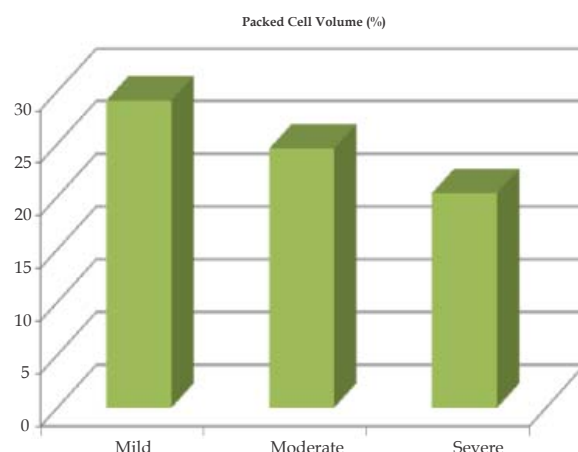


Fig. 3: Association of Packed Cell Volume (PCV) and Severity of Anaemia of females.

Table 3: Association of packed cell volume (PCV) and severity of anaemia of females

Severity of Anaemia	Packed Cell Volume (%)		p-Value
	Mean	SD	
Mild	29.13	2.14	<0.05
Moderate	24.57	1.86	
Severe	20.35	1.12	

Discussion

A hospital based cross sectional study was done among 1000 adult females to evaluate prevalence of spectrum of anaemia. In the present study, majority of the females (42.44%) were in the age group of 26–35 years followed by 31.9% in the age group of 18–25 years, 19.2% in the age group of 36–45 years, 4.9% in the age group of 46–55 years and 1.6% in the age group of 56–65 years. This is similar to the studies of Kumar MR et al, Sharma AK et al, Sarin J et al.³ and Trivedi J et al.⁴

Sarin J et al.³ study determining the prevalence of anemia among antenatal mothers with a view to develop and evaluate a planned health education programme on prevention and management of anemia in pregnancy found (68%) were in the age group of 21–25 years, followed by 21% in the age group of 26–30 years and 10% of the in the age group of below 20 years.

In our study, the mean packed cell volume (PCV) of females with mild anaemia was $29.13 \pm 2.14\%$ while the mean PCV of females with moderate and severe anaemia was $24.57 \pm 1.86\%$ and $20.35 \pm 1.12\%$ respectively. There was significant decrease in packed cell volume (PCV) with increase in severity of anaemia as per ANOVA test ($p < 0.05$). This is in concordance to the study of Sharma AK et al.⁵.

Sharma AK et al.⁷ evaluating the occurrence of Iron deficiency anemia among females of reproductive age group reported majority of women suffering from various grades of anaemia had reduced (77%) red blood count, packed cell volume was also lower than the normal range.

Summary

A hospital based cross sectional study was done among 1000 adult females to evaluate prevalence of spectrum of anaemia. The following observations were noted in our study:

1. Majority of the females (42.44%) were in the age group of 26–35 years followed by 31.9% in the age group of 18–25 years, 19.2% in the

age group of 36–45 years, 4.9% in the age group of 46–55 years and 1.6% in the age group of 56–65 years.

2. 410 (41%) females had mild anaemia while 440 (44%) and 150 (15%) females had moderate and severe anaemia.
3. Microcytic hypochromic anaemia was the most common type of anaemia (56.5%) followed by Normocytic normochromic anaemia (32.4%), dimorphic anaemia (6.3%) and macrocytic anaemia (4.8%).
4. The mean packed cell volume (PCV) of females with mild anaemia was $29.13 \pm 2.14\%$ while the mean PCV of females with moderate and severe anaemia was $24.57 \pm 1.86\%$ and $20.35 \pm 1.12\%$ respectively. There was significant decrease in packed cell volume (PCV) with increase in severity of anaemia as per ANOVA test ($p < 0.05$).

Conclusion

PCV (packed cell volume) and haemoglobin are considered as haematological indicators for classifying the severity of anaemia. Iron deficiency anemia is increasing in females, specially in reproductive age group of developing countries. The haematological parameters can aid in early

recognition of type and cause of anaemia and thereby improve the outcome. Early detection, treatment and prevention of anemia can improve maternal as well as child outcome.

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Stress Levels and its Perceptive Gender Differences Among Generation Z Undergraduate Health Professional Students

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Abstract

Background: Stress experienced by Generation Z (Gen Z) undergraduate health professional students could pose a detrimental effect on their demanding professional lives. Health professional students are known to experience various stressors and it affects their health and academic performance. A gender difference is known to exist in the perception of stress experienced by the students. However, it is not clear if this is true in the case of present-day Gen Z tech-savvy students. Hence, the present study was done to assess the intensity of stress perceived by male and female Gen Z undergraduate health professional students and to evaluate their perception on the role of yoga to overcome it. **Materials and methods:** Descriptive questionnaire-based study was conducted among the Gen Z undergraduate health professional students of a private health university in Dakshina Kannada district of Karnataka, India. One hundred health professional students were administered with a pretested and validated academic stress scale questionnaire. Their stress levels and the responses to various stressors by male and female students were tabulated. Also, their perception on the practice of yoga for alleviating stress was documented. **Results:** Female health professional students are significantly more stressed compared with their male counterparts. Academic stressors such as worrying, failure in formative assessment examination and unable to follow a study time table were significantly more in female students as compared to male students. Stress-related to non-academic stressors such as lack of concentration due to gadget use and lack of courage to speak in the class lecture were significantly more among female students. A significant number of students perceived that yoga is a useful interventional technique in alleviating stress and should be included in their course curriculum. **Conclusion:** There is a female preponderance in the perception of certain academic and non-academic related stressors among Gen Z health professional students. The Gen Z students perceive yoga as a coping strategy to alleviate stress and that it should be incorporated in their course curriculum.

Keywords: Stress; Students; Technology; Yoga.

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Introduction

Stress is defined as 'any challenge to homeostasis' or to the body's internal sense of balance.¹ It is a process by which one can perceive and try to cope up with the demanding environmental threats and challenges.² The sources of stress are called stressors and it disrupts people's daily functioning ability. The healthcare profession is a very demanding

and stressful field and the students undergoing healthcare education are under tremendous pressure to excel in the course work. When stress is perceived negatively it affects not only student's health but also the academic performance.

Academic stress is the response of the body to various academic-related demands that exceed the adaptive capabilities of students.³ Stress perceived by health professional students is dynamic and

dependent on the stage in which the student is in.⁴ The present health professional students belong to Generation Z (Gen Z) who are technology savvy and are good at 'task switch'.⁵ They are digital natives and if they use technology correctly for learning purposes it enhances their participation, engagement and commitment in academics and thus, helping them to perform better in their professional life. Even though students have a positive outlook, attitude and perception towards technology-assisted learning, over usage of digital devices like smart phones, iPad and laptops for non-educational purposes can also lead to detrimental effects causing stress.⁶

Stress is reported to be more prevalent among undergraduate health professional students.⁷ The level of stress perceived by a student depends on the stressful events, socio-cultural factors, gender, ethnicity, educational system, coping processes and personality factors of the individual.⁸ Gender is one of the important determining factors of the stress process. The perception of stress in a given situation and its output i.e. the coping strategies to the stressors is vastly different between the genders.⁹ Increased academic stress in health professional students might lead to various issues like anxiety, anger, depression, low self-confidence, self-esteem, impaired classroom and clinical performance which in turn negatively affect their overall academic performance.¹⁰ Also, elevated corticosteroid levels can impair their memory, concentration and learning ability resulting in negative academic performance.¹¹ This becomes a vicious cycle that can also be detrimental to the student's overall well-being. Hence, the assessment of stress among the Gen Z health professional students and the role of gender in perceiving stress becomes the need of the hour. It will act as a window to the challenges they face and will help in formulating the steps needed to be taken to overcome these hurdles.

Among many coping strategies for stress, practicing Yoga is one of the popular and most effective strategies.¹² Since, there are contradicting reports regarding the perception of stress level among gender,^{11,13,14} we planned to conduct the present study to assess the intensity of stress perceived by male and female Gen Z undergraduate health professional students and to evaluate their perception of the role of yoga to overcome it.

Materials and Methods

A questionnaire-based study was conducted among undergraduate health professional students

of a private health university in Dakshina Kannada district of Karnataka, India. Ethical clearance was obtained from the Institution Ethical Committee (IEC) before the start of the study. Written informed consent was obtained from the study participants after explaining the purpose and nature of the study. Participation was purely voluntary and students were assured that participation would be confidential and would not affect their academic progress.

This cross-sectional study was done on one hundred apparently healthy undergraduate medical and dental health professional student volunteers (purposive sampling) who were born after 1995 (Gen Z).¹⁵ Students who had clinically diagnosed depression, lack of interest, not willing to give written informed consent and incompletely filled questionnaire were excluded from the study.

The questionnaire used in this study is a modified adapted version of the Academic stress scale developed by R. Balaji Rao.¹⁶ It was developed and validated to assess the stress in health professional students. It consisted of 20 questions. The questions were grouped into two main domains such as academic-related stressors and non-academic related stressors. The participant's responses to the questions of the questionnaire were elicited on a Likert scale from 0-4 that most closely described the intensity of stress they perceive. The grading of stress level was as follows; 0-NS (No Stress), 1-SS (Slightly Stressed), 2-MS (Moderately Stressed), 3-HS (Highly Stressed) and 4-ES (Extremely stressed). Total stress score of all the stressors was calculated and they were classified as follows: 1-20 -SS, 21-40 MS, 41-60 HS and 61-80 -ES. They were also asked to respond on their perception of the role of yoga to overcome stress.

Statistical analysis

Data was entered in the MS excel sheet and the data were analyzed using Microsoft excel and the frequency of the stress level among the students was analyzed using SPSS version 16. Descriptive statistics are reported as frequencies and percentages. Chi-square test was used to compare the stress levels associated with various stressors in the male and female students and their perception of yoga to reduce stress. All the tests with $p < 0.05$ were considered as significant.

Results

Among 100 study participants, 68 were female and 32 were male health professional students.

Table 1 shows that female students are significantly more stressed compared to male students.

Stress due to worrying about examination, not

able to keep up with the study time table and failure in internal examination (formative assessment) were significantly more in female students compared to male students (Table 2).

Table 1: Severity of stress among male and female student (N = 100)

Stress level	Male N1 = 32 Frequency (%)	Female N2 = 68 Frequency (%)	p-Value
Slightly stressed	13 (40)	7 (10)	0.002*
Moderately stressed	14 (44)	32 (47)	
Highly stressed	4 (13)	26 (38)	
Extremely stressed	1 (3)	3 (5)	

*Significant (chi-square test)

Table 2: Comparison of academic stressors among the study population (N = 100)

Academic Related Stressor	Males (N1 = 32)					Female (N2 = 68)					p-Value
	0 NS	1 SS	2 MS	3 HS	4 ES	0 NS	1 SS	2 MS	3 HS	4 ES	
<i>Difficult topics</i>	4	11	9	6	2	4	20	21	16	7	0.73
Difficulty in recollecting the studied material	4	9	10	8	1	3	9	21	25	10	0.82
Worrying about the examinations	8	10	7	4	3	4	10	14	21	19	0.003*
Not able to keep up with the study time table	7	9	11	2	3	4	18	20	18	8	0.04*
Apprehensive about results after examinations	9	10	4	5	4	13	10	13	15	17	0.17
Unable to complete the assignment on time	9	11	9	2	1	21	21	17	8	1	0.88
Not able to grasp the subject matter	10	12	6	4	0	9	22	21	12	4	0.12
Last minute preparation for the examinations	7	6	10	6	3	8	15	15	21	9	0.44
Failure in internal examination (formative assessment)	17	2	6	7	0	14	18	9	15	12	0.001*
Fear of becoming additional or repeater	6	9	10	5	7	3	14	25	18	8	0.11

*Significant (chi-square test)

Table 3 shows that lack of concentration during study hours due to use of smart phone or laptop for entertainment purpose and lack of courage to speak in the class were significantly more among female students. Stress due to biased attitude of teacher

was more in both male and female students. It was observed that both male and female participants felt that difficulty in discussing the marks/academic insufficiency with the parents as one of the sources of stress.

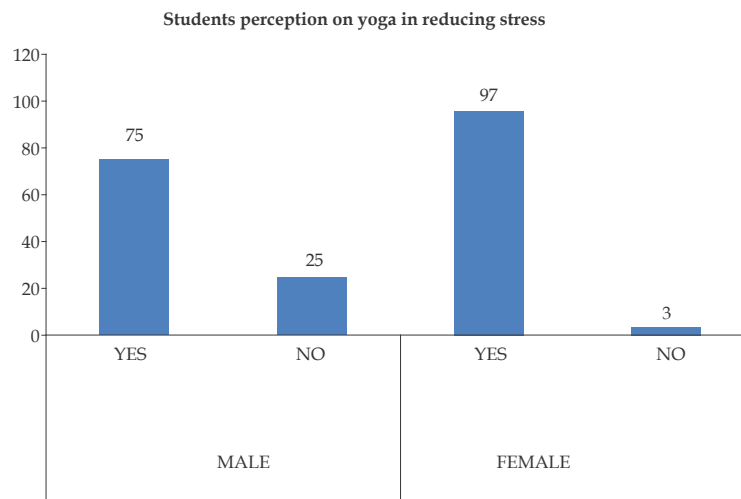
Table 3: Comparison of non-academic related stressors among the study population (N = 100)

Non-Academic Related Stressors	Males (N1 = 32)					Female (N2 = 68)					p-value
	0	1	2	3	4	0	1	2	3	4	
	NS	SS	MS	HS	ES	NS	SS	MS	HS	ES	
Lack of concentration during study hours due to use of smart phone or laptop for entertainment purpose	10	9	4	7	2	10	8	8	13	29	0.003*
Lack of courage to speak in the class	15	10	5	2	0	10	21	18	11	8	0.004*
Not understanding how to prepare for the examinations	9	12	6	3	2	9	25	18	9	7	0.42
Lack of self-confidence	13	8	9	2	0	12	27	20	4	5	0.08
Lack of mutual help among classmates	10	12	6	4	0	22	26	12	5	3	0.72
Lack of fluency while speaking the English language	10	11	6	3	2	23	23	10	6	6	0.98
Biased attitude of teacher	14	9	6	1	2	18	12	22	10	6	0.11
Incomplete and confusing study material	5	14	8	3	2	7	15	36	7	3	0.09
Deficient space or room for study at home/hostel	7	11	5	2	7	14	22	9	8	15	0.94
Difficulty in discussing the marks/academic insufficiency with the parents	14	11	4	3	0	13	16	8	9	5	0.47

*Significant (chi-square test)

Figure 1 shows that significant majority of students (p -value 0.001) perceived that yoga help in reducing stress.

Significant number of the students (p -value < 0.0001) felt that yoga should be included in the curriculum (Fig. 2).

**Fig. 1:** Perception of students on the role of yoga in reducing stress (N = 100).

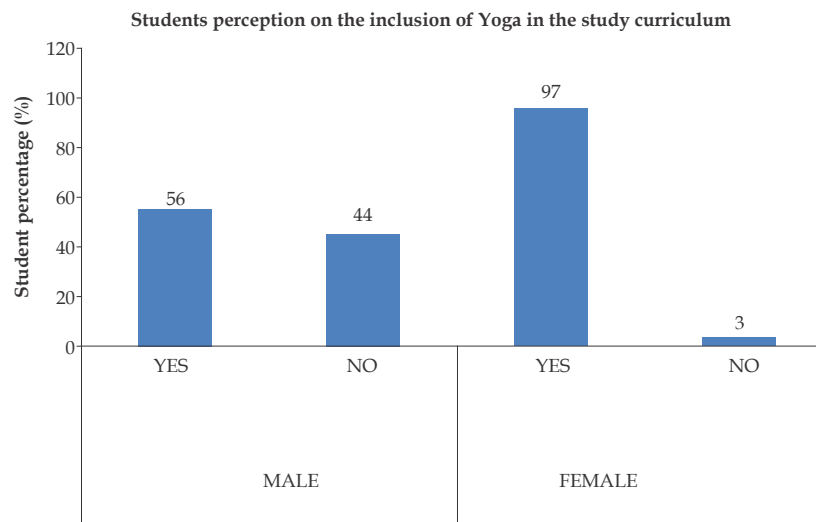


Fig. 2: Perception of students on inclusion of yoga in study curriculum (N = 100)

Discussion

The present-day students are from Gen Z who is finding it difficult to strike a balance between social media, studies and the world around them.¹⁷ In addition, the academic demands and pressure to excel in studies faced by the health professional students are more compared to others. Hence, they are more prone to stress due to various stressors which could eventually lead to mental health issues in them. In addition, the intensity of stress perceived by male and female students of Gen Z students is unclear. Hence, this study was carried out to assess the intensity of stress on Gen Z undergraduate health professional students and the role of gender in perceiving stress. Along with that, the student's perception of the role of yoga to overcome stress was also evaluated.

The findings of our study show that both male and female students pursuing healthcare education have high levels of stress. Iqbal et al. (2015), in his study using the Depression Anxiety Stress Scale, reported that substantial proportions of medical undergraduate students in a college in Bhubaneswar were depressed, anxious and stressed and required immediate professional help.¹⁸ Even though the stress scale used by them is different from ours, the stress levels are found to be predominantly high in undergraduate students in both studies. Stress in health professional students could lead not only to burn out in them but also could lead to a lack of adequate empathy and compromised care towards patients in future.

The comparison of levels of stress depicted that female students were more stressed compared to

their male counterparts. Our study is in accordance with Dahlin et al., and Shah et al.^{19,20} who had also reported a female predominance in relation to stress in contrast to the report by Saxena et al., who had reported that male students were more academically stressed than female students.¹¹ This could be because females are more prone to the symptoms of depression, anxiety, interpersonal sensitivity, somatisation and neurotics compared to males.²¹

The findings of the study also showed that the academic stressors such as worrying about the examination, not able to keep up with the study time table and failure in internal examination (formative assessment) were significantly more in female students compared to male students. Vastness of the academic curriculum, poor performance and increased frequency of examination are few of the common sources of academic stressors in female health professional students.¹⁴ Gen Z students have difficulty concentrating and analyzing complex study concepts which could lead to worrying about and fear of failure in examination.⁵

In our study showed that non-academic stressors such as lack of concentration during study hours due to the use of mobile phones or laptops for entertainment purposes and lack of courage to speak in the class were significantly more among female students compared to male students. This could be because the present-day Gen Z tech-savvy students spend more time with gadgets like mobile phones, laptops, tablet computers and use it for non-academic purposes like browsing the social media platform leading to lack of time for studying and reduced concentration in them. This could

be the result of constant exposure to small bits of information from social media with its six-second patterns of imaging that is rewiring the brain to expect information to be delivered in very short and rapid bursts.⁵ In fact, Patil et al., reported that even though students had positive attitudes about using mobile for learning purposes, given an opportunity they did not show appreciable utilization of mobile for learning.²² Also the social interactions and networking which used to happen in person using emotions and speech figures previously has taken a dramatic U-turn and now happens via social media platforms using emojis and memes. This will ultimately lead to a lack of courage and confidence in expressing themselves in class, especially when they have an audience, therefore, causing stress. Inherently, females are shy by nature and easy availability of expressing their emotions incognito via the social platforms is compounding this nature of female students.²³

It was seen that students perceived a biased attitude of teacher which is one of the teaching-learning stressors as a source of stress. These findings are in contrast with the study reported by Abraham et al., where lesser percentages of students were stressed due to teaching-related stressors.²⁴ A teacher's confidence and unbiased attitude towards the academic potential of a student is very important for their success and also plays a vital role in their academic achievement.²⁵

Our study reported a higher frequency of stress related to stressor such as difficulty in discussing the marks/academic insufficiency with the parents among both genders. The parents of Gen Z students are tired of trying to communicate with their children as they are restless and flighty. Also, both the parents of this generation due to economic demands are working parents who spend more time at work than at home leading to a communication gap between them and their children.²⁶ Hence, the students are unable to connect with their parents and discuss and resolve the academic challenges they face. This compounds to stress faced by both the genders.

Present-day Gen Z health professional students are experiencing an increased amount of stress and need interventional coping strategies to combat the ill effects of stress to excel in their professional life. In our study, the majority of the students are of the opinion that practice of yoga reduces stress and should be incorporated in their course curriculum. Interventional studies were done on health professional students have shown that simple yogic breathing techniques have reduced

the stress levels among the students.^{27,28} Inculcating yoga in health professional study curriculum and knowing its benefits both on maintaining good health and reducing stress will encourage them to practice it and thereby reduce the intensity of both the academic and non-academic stressors.

The specific effects and the correlation of the use of technology on the various individual stressors with their academic performance among Gen Z health professional students could not be assessed and this forms the future scope of the study

Conclusion

Stress experienced by Gen Z health professional students due to various stressors is an important detrimental factor in their professional life. In the present study, females significantly perceived academic-related stressors such as worrying and fear of failing in formative assessment examination and unable to keep up with study time table as more stressful when compared to males. Similarly, non-academic stressors such as lack of concentration because of gadget use and lack of courage to speak in the class were perceived as stressors by females in comparison with their male counterparts. A significant number of students perceived that yoga is a useful interventional technique in alleviating stress and should be included in their course curriculum. Thus, the need of the hour for Gen Z students is for specific targeted measures to substantially reduce the burden of stress in them.

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Airway Resistance in Farmers Exposed to Pesticides as Compared to Controls

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Abstract

Background: Productivity in agricultural field has remarkably increased worldwide in this era of green revolution. Agriculture, a very complex occupation, apart from increasing productivity also brings huge loss to community in terms of its ill effects on humans, plant and animal life due to aggressive use of agrochemicals like fertilizers and pesticides. Respiratory diseases among farmers were one of the first-identified occupational hazards associated with agriculture. As farmers cultivating the earth are the most valuable citizens the present study is designed to detect the ill effects of exposure to fertilizers and pesticides on lung function. **Aims:** Present study was designed to study the effect of pesticides on lung function in pesticide exposed farmers and to compare these lung function parameters in exposed farmers to that of normal controls. **Materials and methods:** Pulmonary function test was studied in 60 apparently healthy male farmers exposed to pesticides, and compared with 62 apparently healthy male control subjects. The pulmonary function test was performed by using an electronic spirometer and results were compared by Student's unpaired *t* test. **Results:** Results of the present study showed a significant decrease in the mean values and percent predicted value of FVC, FEV1, %FEV1/FVC, PEF and FEF 25-75% in pesticide exposed farmers compared to normal subjects. **Conclusion:** Based on results of the present study it may be concluded that pesticide exposed farmers are at increased risk of developing occupationally related pulmonary impairment. There is need for compulsory use of personal protective equipment and measures during pesticide application.

Keywords: Pesticides; Farmers; Pulmonary function.

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Introduction

In the agricultural world, with inadequate rains and insufficient irrigation, there is a persistent crop loss due to pests, diseases and weeds before the harvest.¹

Pesticides are compounds used to save crops from pests attack and to eradicate pests. Usage of these pesticides intensively, leads to its accumulation not only in soil, water, and crops, but also in the surrounding atmosphere.² Pesticides according to their target action includes insecticides, herbicides fungicides and others such as nematicides,

acaricides, rodenticides and fumigants etc. Insecticides contains usually organochlorines, organophosphates, carbamates, pyrethroids, neonicotinoids and phenylpyrazoles. Herbicides contains combination of amides, chlorophenoxy, bipyridyls, dinitroanilines, triazines, urea and amines, phosphonates. Fungicides consisting of inorganic compounds, aromatic, dithiocarbamates, anilides, dicarboximides, (benz) imidazoles and conazoles.³

During agriculture, these pesticides mainly enter the body through skin, followed by respiratory and oral routes.^{4,5}

Inhalation of pesticides occurs during its fumigation, mixture preparation and during application in closed environments.^[6,7] Farmers get exposed to these toxic chemicals due to spills and splashes, direct spray contact as a result of wrong technique of application or inadequate protective equipment's, contaminated shoes, clothes, drinking water and food as well.⁸

These toxic pesticides can accumulate in living species resulting in long-term and chronic effects. Long-term exposure and inhalation of these ultrafine ambient fine particulate matter $\leq 2.5 \mu\text{m}$ (PM_{2.5}) from the agrochemicals pose great adverse effect on respiratory health leading to variety of respiratory diseases like asthma, bronchitis, chronic obstructive lung disease etc.¹

In occupational respiratory diseases, lung function test like spirometry is one of the most important diagnostic tools as it plays a very important role in the diagnosis and prognosis of these diseases and differentiates the effect of restriction or obstruction on the lung function.⁹ Periodic regular lung function testing in farmers can detect pulmonary diseases in its earlier stages where corrective measures are more likely to be beneficial.

Materials and Methods

This study was undertaken in 60 apparently healthy male farmers, aged between 20 to 40 years with history of working for at least 6–8 hours a day for twice a week in the farm. Sixty-two apparently healthy male, control subjects will be selected. Subjects were matched for age, height and weight. Subjects with History of smoking, Alcohol intake, Diabetes mellitus, Hypertension, Anemia, Abnormalities of Vertebral column and thoracic cage, Pulmonary tuberculosis, Bronchial asthma, Chronic bronchitis, other respiratory diseases, Cardiac diseases, Abdominal or chest surgery were excluded from the study.

After taking written consent and asking detailed history about pesticide exposure with series of questionnaires like type of pesticides, mixing procedure, use of protective measures like gloves

and masks, frequency of application etc., Lung function test was performed by using computerized RMS Medspirorin the sitting position at fixed time of the day to minimize the effect of diurnal variation and will be repeated at-least three times after adequate rest. Subjects were motivated prior to the start of the maneuver. Results obtained in computerized spirometer were analysed for Forced vital capacity (FVC), forced expiratory volume in one second (FEV₁), forced expiratory ratio (% FEV₁/FVC), peak expiratory flow rate (PEFR) and forced expiratory flow (FEF 25–75%).

The spirometer is provided with mouthpiece attached to a transducer assembly which is connected to an adaptor box which in turn connected to the computer by a serial cable. Software from recorders and medicare system is loaded onto the computer. The software allows the calculation of the predicted values for age, sex, weight and height and gives the recorded values of all the parameters adjusted for Indian population. The test is simple noninvasive, safe and easy to carry out in the subjects.

Statistical analysis

Was done by using SPSS 20.0 software. Results are presented as mean \pm SD and percentage difference. Unpaired *t* test was used for groupwise comparisons. P value of 0.05 or less was considered for statistical significance.

Results

For the purpose of analysis of data, the results are tabulated as shown in the table. The statistical comparisons of the matching variables (age, height and weight) were inherently similar for both the groups. All lung function parameters are expressed and compared in mean values and percent predicted values adjusted by race, gender, height and age. Farmers exposed to pesticides showed significant reduction in percent predicted values and mean values of FVC, FEV₁, FEV₁/FVC %, FEF₂₅₋₇₅% and PEFR when compared with non exposed controls (Table 1), but these exposed farmers did not show a statistically significant reduction in actual value of FEF₂₅₋₇₅% compared to controls.

Table 1: Lung function data in pesticide exposed farmers compared with their matched controls

Parameters	Controls (<i>n</i> = 40)	Subjects (<i>n</i> = 40)	Significance	
	Mean \pm SD	Mean \pm SD	* <i>t</i> -value	<i>p</i> -value
Age (yrs)	30.92 \pm 6.72	35.13 \pm 6.71	0.01	NS
Height (cm)	1.63 \pm 0.07	1.64 \pm 0.10	0.79	NS

Parameters	Controls (<i>n</i> = 40)	Subjects (<i>n</i> = 40)	Significance	
	Mean \pm SD	Mean \pm SD	* <i>t</i> -value	<i>p</i> -value
Weight (Kg)	61.43 \pm 5.97	57.60 \pm 8.38	0.03	NS
FVC (L)	3.18 \pm 0.41	1.84 \pm 0.29	4.73	<0.001 HS
FEV ₁ (L)	2.73 \pm 0.37	1.69 \pm 0.31	2.93	< 0.05 S
FEV ₁ /FVC (%)	85.71 \pm 2.33	82.34 \pm 4.65	3.89	<0.001 HS
PEFR (L/S)	8.00 \pm 0.91	5.64 \pm 1.17	2.24	< 0.05 S
FEF _{25-75%} (L/S)	4.19 \pm 0.42	3.00 \pm 0.70	1.85	NS

Table 2: Lung function data in (percentage predicted) in pesticide exposed farmers compared with their matched controls

Parameters	Controls (<i>n</i> = 40)	Subjects (<i>n</i> = 40)	Significance	
	Mean \pm SD	Mean \pm SD	* <i>t</i> -value	<i>p</i> -value
FVC (L)	93.05 \pm 4.97	60.40 \pm 6.67	5.84	<0.001HS
FEV ₁ (L)	94.25 \pm 4.05	64.69 \pm 7.35	7.63	<0.001HS
FEV ₁ /FVC (%)	101.35 \pm 3.16	111.18 \pm 9.32	3.89	< 0.05 S
PEFR (L/S)	90.55 \pm 5.83	66.63 \pm 10.50	7.44	<0.001HS
FEF _{25-75%} (L/S)	97.28 \pm 6.46	72.63 \pm 13.34	3.81	< 0.05 S

Discussion

Chronic respiratory diseases affect the entire airway from the ear, nose and throat to the pulmonary alveoli, and are characterised by an inflammatory condition induced or aggravated by viruses, bacteria, fungi and environmental components.^{10,11} Among these, allergens, toxins, tobacco smoke, gaseous or particulate air pollutants and a number of chemicals, including pesticides, can present a risk to the respiratory system.^{12,13,14}

The results of the present study showed that FVC, FEV₁, %FEV₁/FVC and PEFR, FEF_{25-75%} were significantly reduced in the pesticide exposed farmers compared to normal non-exposed subjects which is in agreement with the observations made by other authors.

Mekonnen Y et al. in their study showed that pesticide sprayers had significantly reduced forced expiratory vital capacity (FVC) and forced expiratory volume in 1s (FEV₁) compared to a similarly aged group of nonsprayers.¹⁵

Kesavachandran C et al. and his colleagues in their study found that FVC, FEV₁, PEFR were lower among the pesticide sprayers compared to controls.^[16]

Hernández and his colleagues reported decreased FEF_{25-75%} with lifelong cumulative exposure to pesticides decreased lung volumes.^[17]

Chakraborty S and his co-workers observed decrease in PEFR among the farmers when compared to the control group.¹⁸

While considering the pathophysiological aspects of a decrease in the values of these lung function parameters, FVC is decreased in pulmonary obstruction, emphysema, pleural effusion, pneumothorax, pulmonary edema and poliomyelitis. Similarly, the FEV₁ value is low in obstructive lung diseases and in reduced lung volume.¹⁹ The decline in FEV₁ is an acceptable standard against which we can measure marked declines in subjects with the history of chronic obstructive pulmonary disease (COPD) or in subjects exposed to environmental pollutants, whereas PEFR provides an objective assessment of functional changes associated with environmental and occupational exposures and determines acute or chronic disease processes in patients with severe COPD.²⁰ PEFR is persistently low and represents collapsing of small airways.¹⁹ In view of pathophysiological aspects and a decrease in the lung parameters, our results suggest that exposure to pesticides affects the lung functions. The results of the present study can be attributed to and explained by the following facts.

Pesticides are volatile and become aerosolized, therefore when breathed inside, can cause irritation and subsequent damage to airways resulting in symptoms of chronic bronchitis. Some of the aerosols can reach the alveolar space causing damage to the alveolar sacs leading to breathlessness.²¹ Extended exposure to these irritating toxic substances may cause acute or chronic respiratory ailments.²² Because of the nature of inherent biological reactivity, pesticides are potentially hazardous to human health.²³

The neurological effects caused by cholinesterase inhibiting pesticides especially Organophosphorous compounds and carbamates, are considered to affect airways and lungs, trigger the bronchospasm²⁴ and increase the airflow resistance leading to impaired lung function and respiratory diseases.²⁵ Occupational exposures to pesticides not only affects dynamic lung volumes, may also lead to impairment of gas exchange in the lung.²⁶

Studies have reported that effect of pesticides like Organophosphorous compounds could also be due to the production of oxygen free radicals.²⁷

Several animal studies reported that exposure to Organophosphorous compounds increase the muscarinic effect on airways smooth muscles and the nicotinic effects on muscles of respiration along with increasing affinity to acetylcholine mediating broncho-constriction.²⁸

Conclusion

The present study concluded that pesticide exposure adversely affect the pulmonary function parameters like FVC, FEV1, %FEV1/FVC, PEFR and FEF25–75% in farmers and cause an obstructive pattern of lung function impairment. Were commended to detect respiratory diseases if any, among farmers as early as possible and there is a need to create awareness among the farmers regarding the ill effects of pesticides and to encourage using personal protective equipments and measures, to improve agricultural environments. Farmers can also be educated about the importance of organic farming.

Further studies are needed to particularly focus on effects due to cumulative lifetime exposure to pesticides and identification of specific pesticide exposure, in order to determine possible dose-effect relationships, and thereby assess the causal relationship.

Conflict of Interest: None to declare.

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Glomerular Filtration Barrier and Mechanism of Proteinuria

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Abstract

The first step in urine formation is the filtration of large amounts of fluid through the glomerular capillaries into Bowman's capsule. It is around 180 liters per day. Most of this filtrate is reabsorbed leaving only about 1 liter of fluid to be excreted each day. Glomerular capillary wall (GCW) or glomerular filtration barrier (GFB) were coined during 1950. The GCW consists of three distinct but closely interacting layers: the fenestrated endothelium, with its glycocalyx; the podocytes, with their interdigitated foot processes and slit diaphragm; and the intervening glomerular basement membrane (GBM). Proteinuria is associated with kidney disease and cardiovascular mortality. It can be categorised as glomerular, tubular and overflow proteinuria.

Keywords: Glomerular capillary wall (GCW); Glomerular filtration barrier (GFB); Proteinuria.

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Introduction

The first step in urine formation is the filtration of large amounts of fluid through the glomerular capillaries into Bowman's capsule. It is around 180 liters per day. Most of this filtrate is reabsorbed leaving only about 1 liter of fluid to be excreted each day.¹ Glomerular capillary wall (GCW) or glomerular filtration barrier (GFB) were coined during 1950.²

Glomerular Filtration Barrier/Glomerular Capillary Wall

The GCW consists of three distinct but closely interacting layers: the fenestrated endothelium, with its glycocalyx; the podocytes, with their interdigitated foot processes and slit diaphragms; and the intervening glomerular basement membrane (GBM).¹ Data generated over decades suggest that normal macromolecular filtration requires the contribution of all three layers of the

GFB: Endothelium, GBM and a layer of Podocytes.³

Glomerular Capillary Endothelium

It is fenestrated and has a role in determining protein sieving. The fenestrae are too large to form any meaningful barrier and lack a diaphragm. The luminal surface has glycocalyx forming negatively charged coat that covers the fenestrae with plugs. These are central for charge selectivity. Pre-eclampsia/eclampsia and Diabetic nephropathy are the classic proteinuric diseases associated with endothelial dysfunction.³

Glomerular basement membrane

It is composed of three layers viz. Lamina rara interna (LRI) which consists of sulfated proteoglycans, Lamina densa (LD) consisting of Type IV collagen (COL4) and laminin, and Lamina rara externa (LRE).³

Podocytes with foot process and slit diaphragms

These are highly specialized epithelial cells of mesenchymal origin. Surface of podocytes is covered by anionic glycocalyx, constituted by podocalyxin. Podocytes are characterized by foot processes which interact at specialized cell-to-cell junctions called slit diaphragms.³

Slit diaphragm consist of multiple layers of nephrin strands connecting adjacent foot processes (FPs). Pores within this meshwork form elongated channels. In Nephrin mutation, the slit pores become narrower with shorter, thinner, less organized strands bridging adjacent FPs and forming relatively larger pores and channels. Slit diaphragm is located most distally in the GFB and unlikely to function as restrictive barrier. Proteinuria has also been reported in the absence of FP and SD changes.³

Glomerular sieving coefficient (GSC)

The GSC is the ratio of a molecule concentration in Bowman's space to that in plasma. As albumin is the most abundant plasma protein, albuminuria is one of the most important signs of glomerular disease. Determining albumin's GSC is central to understanding the GFB.⁴

Controversies regarding glomerular filtration barrier

The controversies related to glomerular filtration barrier revolves around (1) the size verses charge selectivity (2) the Slit Diaphragm verses Glomerular Basement Membrane as predominant albumin barrier and (3) the Glomerular barrier verses Tubular reabsorption.⁴

Size vs. charge selectivity

The existence of size selectivity is universally accepted. Studies done with inert tracers such as Ficoll and dextran indicate that the Glomerular Sieving Coefficient (GSC) is inversely related to molecular weight and radius. Negatively charged dextran was more restricted than neutral dextran, which was more restricted than positively charged dextran of similar size⁴

Charge selectivity is not universally accepted and has been challenged by different workers. As

reducing fixed anionic charge sites in the GBM by more than 50% has no consequences on urinary albumin concentration⁴.

Slit Diaphragm vs. Glomerular Basement Membrane

The interaction between $\alpha 3$ - $\beta 1$ integrin and $\beta 2$ laminin is important for normal podocyte-GBM function. Activation of $\alpha 3$ - $\beta 1$ integrin results in recruitment of a integrin-linked kinase (ILK). When ILK is deleted in a podocyte the proteinuria developed with foot process effacement and glomerulosclerosis⁴

On the other hand, blocking (Transforming Growth Factor) TGF- $\beta 1$ or adriamycin induced activation of ILK preserved podocyte and ameliorated albuminuria. Together, these findings suggest that the regulation of $\alpha 3$ - $\beta 1$ integrin/ILK may be important in optimal podocyte and GFB function⁵.

Glomerular barrier vs. Tubular reabsorption

Mechanism of protein filtration

As per "Gel Permeation/Diffusion Hypothesis" laid down by Oliver Smithies, diffusion through the GBM, is the predominant force governing macromolecular movement through the GFB. Diffusion is independent of fluid flow (i.e., GFR), but dependent on the gel's properties. According to this hypothesis, increased protein concentration in the glomerular filtrate can occur by two different pathways.⁶

The first is by an increase in the rate of passage of protein by changes in the gel's properties (i.e. by alterations to the GBM and perhaps also to the endothelial glycocalyx).⁶

The second is by reduction in the available surface area for filtration, as occurs either with FP effacement or reduced endothelial fenestration. The increased protein concentration in the filtrate then overwhelms the tubular reabsorption capacity, resulting in albuminuria.⁵

Role of tubular absorption

There is significant post glomerular processing of albumin by tubules. 94% of the filtered albumin is absorbed by proximal tubules. Studies concluded that proteinuria is a tubular rather than a glomerular disorder⁵.

Proteinuria

Urinary protein excretion in the normal adult humans is less than 150 mg/day⁷. Proteinuria is associated with kidney disease and cardiovascular mortality⁸.

It can be Glomerular due to impairment of the glomerular filtration apparatus, tubular due to diminished tubular reabsorption of low molecular weight proteins and overflow when large loads of filtered proteins exceeds resorptive capacity⁵.

Glomerular proteinuria can be categorized as Non pathological and Pathological. The Non pathological proteinuria is further divided into transient and orthostatic.⁵

Transient proteinuria occurs in patients with normal renal function with bland urine sediment and normal blood pressure. The quantitative protein excretion is less than 1 g/day. The proteinuria is not indicative of significant underlying renal disease. This is precipitated by high fever or heavy exercise.⁹

Orthostatic proteinuria means no proteinuria in early morning samples but low-grade proteinuria at the end of the day. Usually occurs in tall, thin adolescents or adults younger than 30 years. Patients have normal renal function and proteinuria usually is less than 1 g/day with no hematuria⁵.

Pathological proteinuria can be categorized by protein quantity into Non-nephrotic proteinuria i.e. proteinuria <3.5 g/24 h and Nephrotic proteinuria i.e. proteinuria >3.5 g/ 24 h.⁵

Tubular proteinuria occurs most commonly in disease processes affecting the tubulo-interstitial component of the kidney. It comprises low molecular weight proteins such as beta-2 microglobulin, which in normal conditions are completely reabsorbed by proximal tubules. The amount of proteinuria is <2 g/ 24 h and dipstick test may be negative⁵.

Overflow proteinuria is associated with increased production of abnormal low molecular weight proteins (eg. light chains in multiple myeloma, myoglobin in rhabdomyolysis) that exceeds the reabsorption capacity of the tubules, leading to spilling of the protein into the urine. These low molecular weight proteins can be toxic to the tubules and can cause acute kidney injury.⁵

Conclusion

It is concluded that the GFB as a functional unit consists of three different elements. It is suggested that it should be viewed as a dynamic sieve, rather than as physically inert. Normal GFB function requires not only three intact GCW layers, but also a hemodynamic steady state in the glomerular capillary and the urinary spaces. A change in any factor (GBM, cell, glycocalyx, local GFR, or plasma albumin concentration) will alter albumin's concentration in the primary filtrate. When the level of albumin in the filtrate exceeds the tubular absorption threshold, albuminuria will ensue.

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