Original Research Article

Assessment of clinical significance of serum uric acid, serum bilirubin and serum creatinine in patients with breast lump

Shoranki Pardhan*, Anuradha Chaudhary, Rahul Shivhare, Sonveer Gautam

Department of General Surgery, Gandhi Medical College, Bhopal, Madhya Pradesh, India

*Correspondence:
Dr. Shoranki Pardhan,
E-mail: shoranki14394@gmail.com

ABSTRACT

Background: Breast cancer is the most common cancer in females, approximately a quarter of all cancers. Several tumor markers are being studied now a days like α feto protein, carcinoembryonic antigen, and human chorionic gonadotropin but, the cost of these analytical methods is very high. This study is directed to assess prognostic value of serum uric acid, serum bilirubin and serum creatinine in breast lump cases. And to compare variation in benign and malignant breast lumps in relation to serum uric acid, serum bilirubin and serum creatinine values.
Methods: Observational and prospective study conducted in Hamidia Hospital, Bhopal from November 2018-July 2020 included 100 patients. 50 patients were having benign breast lumps and other 50 patients malignant. Categorization of the patients done on the basis of histopathological examination. Autoanalyzer was used for estimation of serum uric acid, serum bilirubin and serum creatinine.
Results: High serum uric acid level may anticipate poor prognosis in carcinoma breast, and it may perform a promising role in the management of the same. The level of serum bilirubin was slightly more or within normal range. The level of serum creatinine was within normal range in absence of any renal impairment by other etiology.
Conclusions: The evaluation of serum uric acid and serum bilirubin may be useful for predicting prognosis in females with malignant breast lump and can provide help in making diagnosis during the initial assessment of the patient presenting with breast lump.

Keywords: Biochemical parameters, Breast cancer, Prognosis of carcinoma breast, Anti-oxidant property

INTRODUCTION

Breast is an actively advancing structure which transits through the different phases of female reproductive life, coinciding with recurrent variations during menstrual cycle. The pathogenesis of breast lump involves disturbance in breast physiology extending from an extreme normality to well defined disease process summarized as aberrations of normal development and involution (ANDI). Self-breast examination by the patient, in fact, discovers 85 % or more of breast lumps.[1,2] Breast lump become palpable when it is more than 1 cm in diameter. It takes 3 years for a tumor to attain this size from a single cell stage estimated by tumor doubling time.[3]

Breast cancer has now ranked first in cancer among Indian females with rate as high as 25.8 per 100,000 females and mortality rate 12.7 per 100,000 females.[4] Earlier cervical cancer was the most common cancer in Indian females but over the last 20 years or so the incidence of breast cancer has transcended cervical cancer and is the leading cause of cancer death.[5]

However, in rural India cervical cancer still remains the common one.[5] According to Globocan 2018; India, United states and China collectively accounts for approximately 1/3 of global breast cancer burden. India is facing crucial condition due to 14% increase in incidence and 11.1% in mortality caused by breast cancer (2012-2018).

The main problem from patient's point of view is apprehension that such a lump may be a cancer. Most of the time breast lump is benign (non-cancerous) but can be malignant (cancerous) and can be proved so by histopathological examination. Breast cancer is the most common cancer in females worldwide, contributing approximately a quarter (25%) of all cancers. Number of cases of breast cancer in developing countries is relatively higher than the developed countries.[6]

The clinician should always provide some level of counselling to the patient after making provisional diagnosis through history taking and physical examination. He/she should resolve all queries and doubts of the patient keeping in mind the possibility of the tumor to be malignant and also be providing some degree of diagnostic accuracy while at the same time ensuring that an excessive biopsy rate is prevented.

It has now become much easier to rule out malignancy with the development of diagnostic aids such as mammography, ultrasonography and aspiration cytology. All of these provide benefit of being non-invasive. Speaking of non-invasiveness, recent studies have shown prognostic significance of some biochemical parameters in cases of breast lump.

Several tumor markers are being studied now a days, which include alpha-feto-protein (AFP), carcinoembryonic antigen (CEA), human chorionic gonadotrophin (HCG), prostate specific antigen (PSA) and comparatively lesser specific markers like lactate dehydrogenase (LDH), alkaline phosphatase (ALP), ferritin and gamma glutamyl transpeptidase (GGT) and reduced glutathione.

Although, the analytical method of some of the above-mentioned parameters are not approachable for common population as these investigations are only available at well-equipped sophisticated centers having newest technology and are also expensive.

And hence therefore, medical practitioners need some simple biochemical parameters to detect possibility of any malignancy which can be assayed easily and are less expensive. And those are serum uric acid, serum bilirubin, serum albumin, serum creatinine however, all of these are having relatively less specificity. However, elevated level of serum uric acid is postulated to be associated with poor prognosis of the breast cancer patient coinciding with ER and PR negative status.

The biochemical parameters focused in this study are: serum uric acid, serum bilirubin and serum creatinine. This study was directed to assess prognostic value of above-mentioned parameters in the patients of breast lump. And since the present study includes all breast lumps whether it is benign or malignant, it will also perform major role in comparing the variation between benign and malignant breast lumps concerning with aforementioned parameters.

Objectives

Objectives of the research were: to study prognostic significance of serum uric acid, serum creatinine and serum bilirubin in females presenting with the breast lump; to study variation in clinicopathology in benign and malignant breast lumps in relation to serum uric acid, serum creatinine and serum bilirubin; and to study relation of serum uric acid, serum bilirubin and serum creatinine with receptor status (ER, PR and HER-2neu), BIRADS score, stage of the tumor and grade of the tumor.

METHODS

The design of the present study was observational and prospective, conducted in the period November 2018 to July 2020. Approval of institutional ethics committee was provided as letter no. 36160-62 dated 14 November 2018.

Study conducted on all the outpatient and inpatient diagnosed to have a breast lump, admitted in all the units of department of general surgery, Gandhi Medical College and Hamidia Hospital, Bhopal. All the patients presented to surgery outpatient clinic with the features suggestive of breast lump were subjected to detailed history and clinical examination. Standard protocol for triple assessment was followed. Proper consent was also taken.

Around 5 ml of venous blood was collected from antecubital vein under aseptic precautionary measures using sterile disposable syringe. Blood was then allowed to clot and serum was separated by centrifugation and stored for testing.

Total 100 patients were included in this study (sample size-100), out of which 50 patients were having benign breast lumps and other 50 patients were having malignant breast lumps. Sample size was calculated by using online sample size calculator, for which confidence level was taken 95%, population size 1000 (it is the number of breast lump patients assessed during the period of study) and margin of error 9%.

Categorization of the patients was done on the basis of their histopathological examination report. Radiological findings were also recorded in all the patients taking part in this study.

In the patients with surgical intervention and in whom surgery was contemplated underwent excisional biopsy all were subjected to histopathological examination of the lump.

Breast lumps proved to be benign by fine needle aspiration cytology (FNAC)/biopsy were not subjected to further histopathological analysis (ER/PR and HER2/neu status). Breast lumps proved to be malignant by histopathological examination then further subjected to analysis of ER/PR status and HER2/neu status.

Inclusion criteria

The study included only females with 16 years of age and onwards.

Exclusion criteria

Female patients who have not attained menarche with history of any trauma to the breast and patients not giving consent for any sort of surgical intervention were excluded from the study.

Methodology

Blood samples (venous blood, 5 ml) of the patients were taken and processed under all aseptic precautions. Autoanalyzer was used for estimation of above-mentioned biochemical parameters by biochemistry technician in central pathology laboratory (CPL) of Hamidia Hospital, Bhopal. The autoanalyzer machine was based on the principle of spectrophotometry. The automated clinical chemistry analyzer used for this study was Biosystems BA 400. It incorporates optical LEDs based system with 8 working wavelengths (340, 405, 505, 535, 560, 600, 635, 670 nm) for spectrophotometric readings, having photometric range of -0.2 to 3.5 A. For present study, normal range for serum uric acid is 3 mg/dl to 5 mg/dl (standardized as per our laboratory report).

Statistical analysis

Data collected were transformed into variables, coded and entered in Microsoft excel. Data was analyzed and statistically evaluated using statistical process control calculator run on a personal computer (SPCC-PC-21 version). Quantitative data was expressed in mean±standard deviation and depends on normality distribution difference between two comparable groups were tested by student's t-test (unpaired) or Mann Whitney 'U' test while for more than two groups analysis of variance (ANOVA) test or Kruskal Wallis H test followed by posthoc test was used. Qualitative data were expressed in percentage.

Statistical differences between the proportions were tested by chi square test or Fisher's exact test. Spearman correlation coefficient was used to see the correlation between two quantitative variables. 'P' value less than 0.05 was considered statistically significant.

Statistical differences between the proportions were tested by chi square test or Fisher's exact test. P value <0.05 was considered statistically significant and p value <0.01 was considered highly significant.

RESULTS

The association between various parameters shown in the graphs above were having equivocal findings. No significant correlations were found after comparing HER 2/neu positive and HER 2/neu negative (Figure 3); ER/PR positive and ER/PR negative (Figure 4); and various stages of breast cancer (Figure 5).

On comparing, positive correlation was found between BIRADS score and serum uric acid and serum bilirubin as shown in Table 1.

P value was found to be highly significant statistically.

Table 1: Association of BIRADS score with biochemical parameters in breast tumours (n=100).

BIRADS score	N	Mean	P value
Serum uric acid (mg/dl)			
1	15	4.24±0.53	<0.001
2/3	37	4.31±0.82	
4a	8	6.77±0.18	
4b	19	6.92±0.33	
4c	14	6.79±0.48	
5	7	6.87±0.40	
Serum bilirubin (mg/dl)			
1	15	0.90±0.19	<0.001
2/3	37	0.95±0.22	
4a	8	1.28±0.17	
4b	19	1.38±0.20	
4c	14	1.49±0.34	
5	7	1.50±0.30	
Direct bilirubin (mg/dl)			
1	15	0.29±0.11	<0.001
2/3	37	0.28±0.13	
4a	8	0.50±0.05	
4b	19	0.56±0.09	
4c	14	0.57±0.16	
5	7	0.63±0.17	
Indirect bilirubin (mg/dl)			
1	15	0.60±0.14	<0.001
2/3	37	0.67±0.13	
4a	8	0.78±0.15	
4b	19	0.82±0.16	
4c	14	0.92±0.22	
5	7	0.87±0.25	
Serum creatinine (mg/dl)			
1	15	0.95±0.18	0.10
2/3	37	0.85±0.17	
4a	8	0.86±0.14	
4b	19	0.90±0.15	
4c	14	0.98±0.19	
5	7	0.84±0.19	

Although, no significant correlation was found between biochemical parameters used in this study (serum uric acid, serum bilirubin and serum creatinine) and age, BIRADS score, stage of tumor, tumor grade as shown in Table 2.
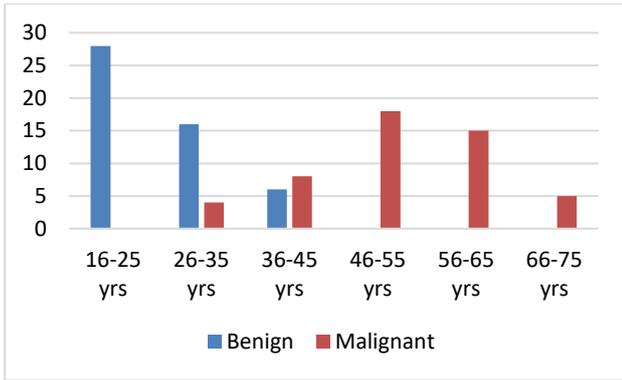
Figure 1: Age wise distribution of breast lumps.
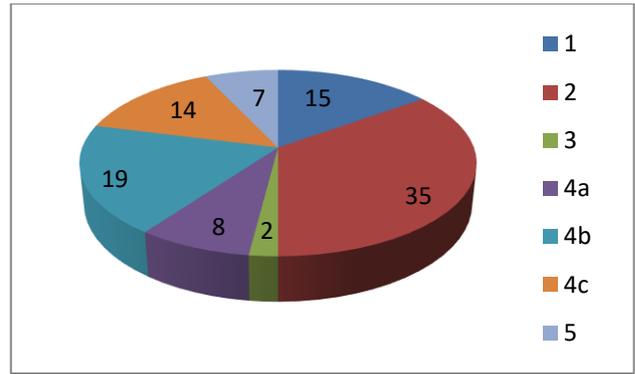
**Figure 2: BIRADS score of distribution in study subjects (n=100).**
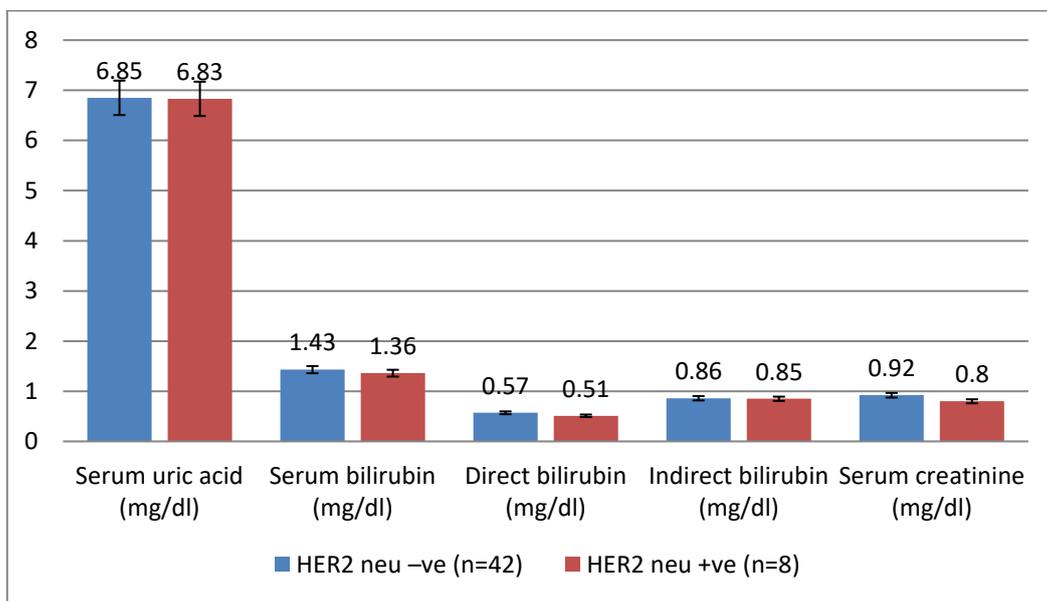
Figure 3: Association of HER2neu status with biochemical parameters in malignant tumours (n=50).
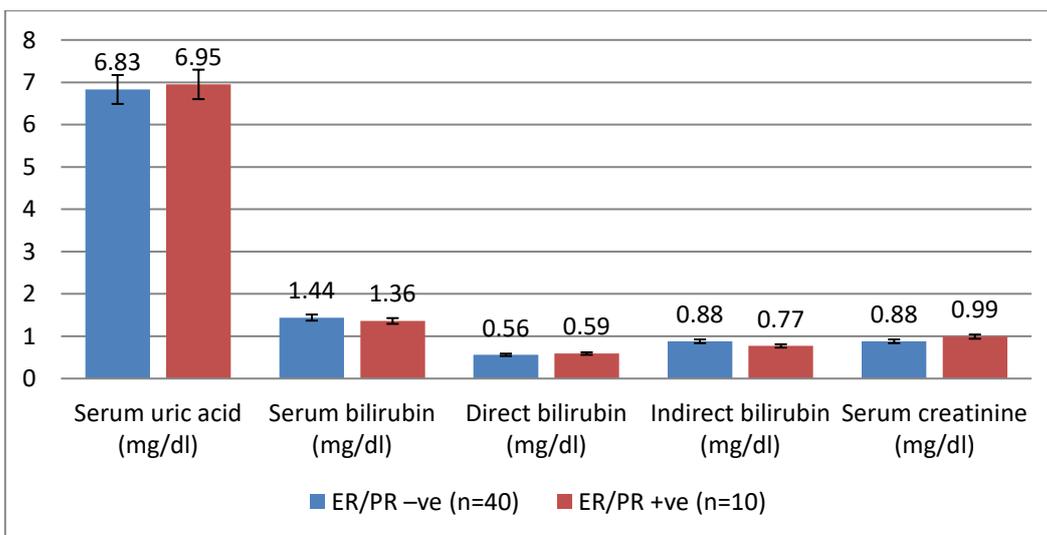

Figure 4: Association of ER/PR status with biochemical parameters in malignant tumours (n=50).
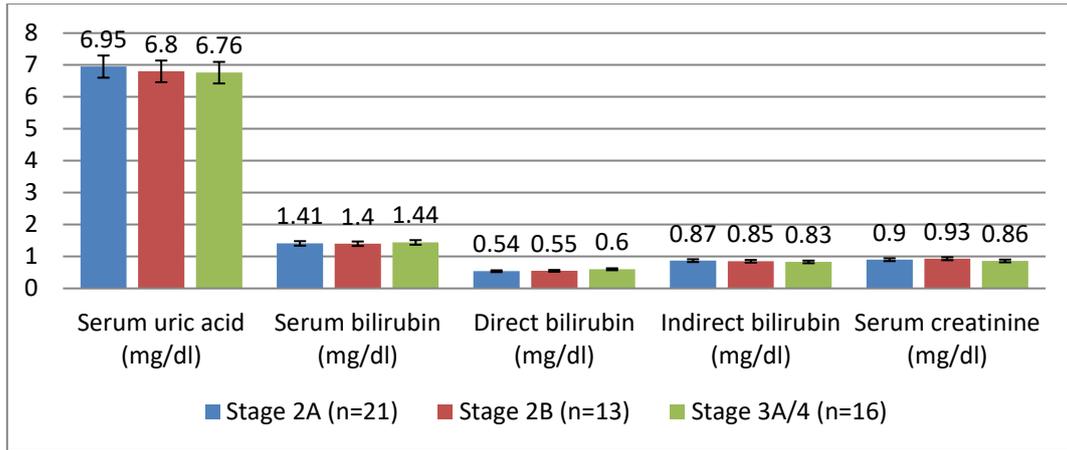
Figure 5: Association of stage of tumour with biochemical parameters in malignant tumours (n=50).

Table 2: Correlation of biochemical parameters with age, BIRADS score, stage and grading.

Biochemical parameters	Age in years	BIRADS score	Stage	Tumour grade
Serum uric acid (mg/dl)				
R value	0.714	0.768	-0.148	0.034
P value	0.000	0.000	0.306	0.812
N	100	100	50	50
Serum bilirubin (mg/dl)				
R value	0.692	0.727	0.074	-0.027
P value	0.000	0.000	0.609	0.853
N	100	100	50	50
Direct bilirubin (mg/dl)				
R value	0.750	0.727	0.234	0.006
P value	0.000	0.000	0.102	0.966
N	100	100	50	50
Indirect bilirubin (mg/dl)				
R value	0.422	0.514	-0.059	-0.083
P value	0.000	0.000	0.684	0.567
N	100	100	50	50
Serum creatinine (mg/dl)				
R value	0.043	0.001	-0.097	-0.002
P value	0.672	0.994	0.504	0.992
N	100	100	50	50
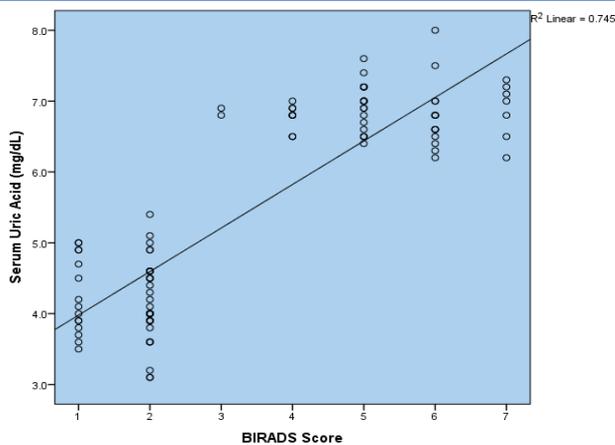
Figure 6: Scatterplot showing correlation between serum uric acid and BIRADS score.
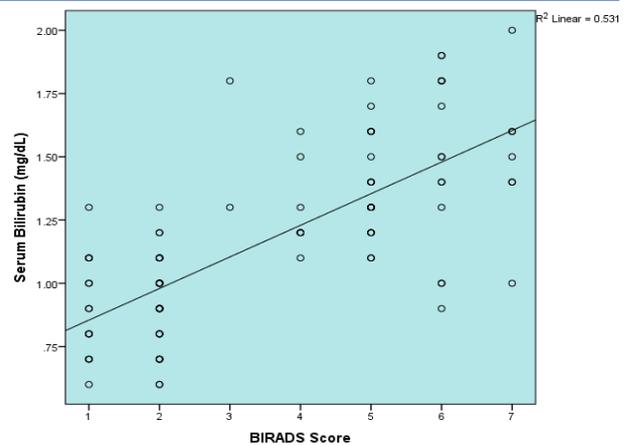
Figure 7: Scatterplot showing correlation between serum bilirubin and BIRADS score.

DISCUSSION

Complete blood picture is a prerequisite investigation for breast lump patients before use of any sort of treatment guideline. Blood chemistry panel is common tests used to evaluate a variety of chemical components excreted from body tissues or synthesized during the breakdown or metabolism of various substances. The blood chemistry panel measures the levels of chemicals, enzymes, and organic waste products detected in the blood stream. It evaluates the healthiness and proper functioning of various organs during the course of disease. The abnormal blood chemistry report also suggests the spread of breast cancer to the bone, kidney or liver.

Some of the studies have investigated the associations of liver function tests (LFTs) and renal function tests (RFTs) with prognosis and mortality in breast cancer.[7] In this present study, the functioning of liver and kidney were assessed to check the level of different components. The LFT measured the level of important chemicals including serum bilirubin (direct and indirect bilirubin). The two forms bilirubin were measured to determine the conditions such as liver disease, hemolytic anemia, and blockage of the bile ducts if any. The RFT is used to estimate values of various elements including uric acid and creatinine in the blood stream which is performed to evaluate the kidney functioning.

In this present study conducted in Hamidia Hospital, serum uric acid levels in patients with malignant breast lump (n=50) was raised then the patients with benign breast lumps (n=50; can be considered as control group) which is close to the result reported by several other studies, and manifested that serum uric acid level may be a protective agent and its role as antioxidant and raised serum uric acid levels as risk factor for incidence and cancer mortality in breast cancer of females.[8-11]

According to Krishna et al, a significant increase in uric acid levels in untreated females with breast cancer, which may be due to high oxidative stress.[9] In our study, level of serum bilirubin in breast cancer patients was found to be slightly raised than normal limits but when compared with bilirubin level of benign breast lump patient, it showed insignificant correlation. However, some studies reported correlation of serum bilirubin and oxidative stress due to carcinogenesis in females with breast cancer.[11]

Antioxidant like uric acid and bilirubin may be important in determining the oxidant and antioxidant status in female patients with breast cancer. In the etiology of cancer, involvement of oxidant such as hydrogen peroxide (H_2O_2), singlet oxygen ($1/2\ O_2$) and superoxide anion (O_2) and hydroxyl radicals (OH) are significant and well recorded in previous studies.[12] It has already been suggested that endogenous antioxidants, explicitly serum bilirubin and serum uric acid, can perform a major role against carcinogenesis due to their anti-carcinogenic properties.[13-16] Pertaining to serum uric acid and serum bilirubin, ascertainment from prognosis studies is less accordant, with some seeking better outcomes in cancer patients with higher blood levels while opposite is signified by others.[17-25]

Regarding serum uric acid, it was displayed by a recent meta-analysis based on nine prospective studies that there is a direct correlation with overall cancer mortality, although notable heterogeneity of findings from the included studies was reported.[26]
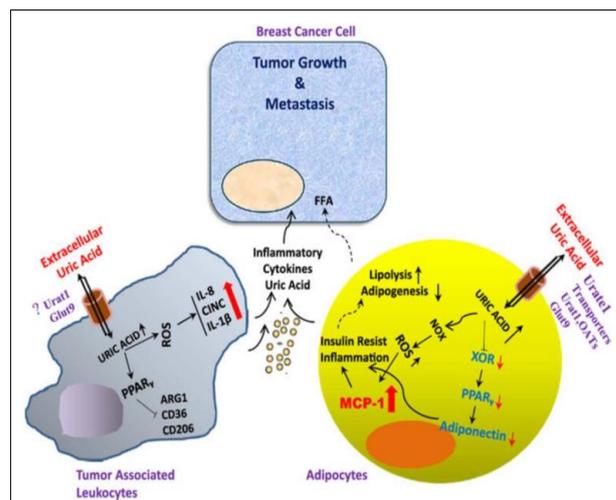

Figure 8: Pro-tumorigenic role of uric acid in breast cancer microenvironment.[27]

It has been postulated previously that oxidative stress contributes to carcinogenesis. Bilirubin is a potent antioxidant and has been shown to protect against cancer development. Lower serum bilirubin level has been associated to raised risk of cardiovascular disease and cancer.[28,29] The association of serum bilirubin level with survival has been evaluated in metastatic breast cancer patients and the results showed that hyperbilirubinemia was related with worse survival.[30] The level of bilirubin in blood stream may have different predictive effect in metastatic and non-metastatic breast cancer patients. In our study, serum bilirubin level in breast cancer patients were found to be slightly more than normal range. This may be due to bilirubin scavenge during oxidative load or oxidative stress in breast cancer.

Limitations

The present study had many limitations. Blood biochemistry parameters were only available from one time point. However, it is unlikely that the routine biochemical parameters analyzed in the present study undergo substantial intra-individual changes overtime. Nevertheless, we have to acknowledge that relation between bilirubin and uric acid with cancer risk in epidemiological studies have been heterogenous, and that stronger experimental evidence is needed to corroborate

hypotheses on potential anti-carcinogenic properties of bilirubin and uric acid.

CONCLUSION

The present study focused on evaluation of serum biochemical parameters (uric acid, bilirubin and creatinine) in patients with breast lump (50 patients with benign lump and 50 patients with malignant lump). In this study, we concluded that raised level of serum uric acid may be due to its protective role in response to increased oxidative stress and high serum uric acid level may suggest poor prognosis in patients with breast cancer. And thus, it may play a major role in providing adequate management for patients with breast cancer. The level of bilirubin was slightly higher than normal range or within normal range, it could be due to bilirubin scavenge during oxidative load or oxidative stress in female patients with breast cancer. The level of serum creatinine was within normal range for both benign and malignant breast lump patients in absence of any renal impairment by other etiology. The evaluation of patient's biochemical profile along with abovementioned parameters, may be used as a credible predictive mean in the monitoring of disease, metastasis and for different treatment modalities for breast cancer. These results suggest that evaluation of serum uric acid and serum bilirubin may be useful for predicting prognosis in females with malignant breast lump (breast cancer) and can provide help in making diagnosis during the initial assessment of the patient presenting with breast lump.

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