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ORIGINAL ARTICLE

Procalcitonin as a predictor of severity and mortality in a cohort of patients hospitalised with COVID-19

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Abstract

Introduction: To evaluate the association of Procalcitonin (PCT) with severity in Coronavirus disease 2019 (COVID-19), hospitalised patients and to test the hypothesis that it is an independent predictor of mortality. *Materials and Methods:* This study was conducted at Chemical Pathology, Department of Pathology and Laboratory Medicine and Department of Medicine, Aga Khan University (AKU), Karachi Pakistan. Electronic medical records of all in-patients including both genders and all age groups with documented COVID-19 from March to August 2020 were reviewed and recorded on a pre-structured performa. The subjects were divided into two categories severe and non-severe COVID-19; and survivors and non-survivors. Between-group differences were tested using the Chi-square and Mann–Whitney's U-test. The receiver operating characteristic curve was plotted for serum PCT with severity and mortality. A binary logistic regression was used to identify variables independently associated with mortality. The data was analysed using SPSS. *Results:* 336 patients were reviewed as declared COVID-19 positive during the study duration, and 136 were included in the final analysis including 101 males and 35 females. A statistically significant difference in PCT was found between severe and non-severe COVID-19 (p value=0.01); and survivors and non-survivors (p value<0.0001). PCT, older age and increased duration of hospital stay were revealed as variables independently associated with mortality. On ROC analysis, an AUC of 0.76 for mortality prediction was generated for PCT. *Conclusion:* Baseline serum PCT concentration is a promising predictor of mortality and severity in COVID-19 cases when considered in combination with clinical details and other laboratory tests.

Keywords: Procalcitonin, COVID-19, severe, mortality, prognosis

INTRODUCTION

Wuhan, China, emerged as the epicenter, of the global pandemic of novel coronavirus disease 2019 (COVID-19) caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), with genetic proximity closer to SARS-CoV and Middle East respiratory syndrome coronavirus (MERS-CoV) in 2019.[1,2] The outbreak led to serious economic, social, medical and psychological implications.[3] Confirmatory diagnostic testing based on molecular platforms was utilised as the gold standard.[4] However, the mysterious presentation, dearth of reliable scientific literature on the subject, lack of evidence on the use of prognostic biomarkers, and the complexity of the disease accompanied by a rapidly evolving clinical course, kept the medical fraternity perplexed globally.[5]

Smart resource allocation emerged as a key element for the already overwhelmed health care systems globally.[6] Intensive care provision can be driven by biomarkers of risk stratification, and there remained an urgent and extensive need for scientific evidence on the utility of biochemical markers related to COVID-19 disease severity and mortality.[7] As the pathogenesis of this novel infection included a plethora of inflammatory cascades, cytokine storms, and the activation of

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coagulation pathways. These triggers may lead to systemic vasculitis (pulmonary, renal, and cerebral), progressing to systematic inflammatory syndrome and even sepsis in most cases.[8] One biomarker of particular focus is PCT, owing to the characteristic elevation in bacterial compared to viral infections and widely established capability as a prognostic biomarker and guide for antibiotic stewardship in various inflammatory states.[9]

PCT is classified as a glycoprotein, which in a normal state is synthesised by the CALC-1 genes in the C cells of the thyroid gland, however, in case of microbial invasion, calcitonin gene expression leads to its release from all parenchymal tissues.[10] Various studies have shown that healthy subjects without any exposure to infectious agents have serum PCT levels below 0.05 ug/L.[11] Cytokines, such as interferon (INF)-γ, which are released following viral infection, lead to down-regulation of PCT, a point worth considering while evaluating the prognostic utility in COVID-19.[7] PCT follows rapid kinetics and rises within 2–6 h after the stimulus, however, contrasting opinion exists on its utility as a prognostic marker for COVID-19 patients.[12,13,14] Given these unique characteristics and the potential association with severity, the aim of this study is to evaluate the association of PCT with severity in COVID-19 in-patients and to test the hypothesis that it is an independent predictor of mortality.

MATERIAL AND METHODS

This retrospective observational study was conducted at the section of Chemical Pathology, Department of Pathology and Laboratory Medicine, in collaboration with the Molecular Pathology and Intensive Care Unit, Department of Internal Medicine, Aga Khan University (AKU), Karachi after seeking exemption from Ethical Review Committee of AKU. The centre is a tertiary care non-profit hospital, situated in the economic and financial hub, largest and the most densely populated city of Karachi, Pakistan. The hospital is the largest medical facility in Pakistan with many specialised subspecialty medical services. It is accredited by the Joint Commission International (JCI) and the laboratory services are accredited by JCIA and College of American Pathologists (CAP) accredited. Being a national referral centre, the population being served is representative of the entire geographical distribution of the country.

Medical records of all in-patients including both genders and all age groups with SARS-CoV-2 positive on a reverse-transcriptase Polymerase Chain Reaction (PCR) test of a specimen collected on a nasopharyngeal swab using Cobas® SARS-CoV-2 Qualitative assay for use on the Cobas® 6800/8800 Systems (Roche Molecular Systems) from 1st March to 10th August 2020 were reviewed. Only laboratory-confirmed cases were included and studied in detail. Patients who remained admitted in the hospital at the time of data compiling on 10th August 2020 were not included as their outcomes were not known.

The demographic details, length of stay in the hospital, and outcome (survived or expired) at the time of discharge along with the results of baseline PCT within 24 hours of hospital admission were recorded on a pre-structured questionnaire. The study was approved by the institutional ethical review committee of the AKU, Karachi (ERC#2020-5168-14099). The subjects were divided into two groups having non-severe and severe COVID-19. Severity was defined as having either of the two criteria; requiring intensive care admission or assisted respiration based on invasive ventilation or non-invasive oxygen support. For assessing the role of PCT in mortality prediction, the study sample was further divided into two groups i.e. survivors and non-survivors.

Serum PCT was measured by Electro-Chemiluminescence immunoassay (ECLIA) on the Roche Elecsys E411 immunoassay analyzer using the manufacturer's recommendations. Results are expressed as microgram of PCT per litre of serum (ug/L). For internal quality control, 2 levels of manufacturer-provided controls (low and high) were run with each batch of samples whilst samples from CAP were run for external quality control during the study period and were found acceptable.

The continuous variables with normal distribution were expressed as mean ± 2 standard deviations, whereas continuous variables with an asymmetric distribution were expressed as median and the respective interquartile range (IQR). Shapiro Wilk test was used to check the normality of PCT data. As the data was skewed; median values were reported along with interquartile ranges (IQR) for PCT. Between-group differences will be tested using the Chi-square test for categorical data and Mann–Whitney's U-test for continuous. $p < 0.05$ will be considered statistically significant and $P < 0.01$ as highly significant. The receiver operating characteristic curve (ROC) was plotted

to further evaluate the relationship of PCT with severity and mortality respectively and the area under the curve (AUC) calculated. A binary logistic regression was used to identify variables independently associated with mortality. The data was analysed using Statistical Package for the Social Sciences version 26 (IBM Corp., Armonk, NY).

RESULTS

A total of 336 in-patients were reviewed as declared COVID-19 positive as per positive PCR results during the study duration. 97 (29%) cases were excluded in the initial scrutiny as they did not continue their treatment at AKU and hence further workup was not conducted. The study sample was further dissected and only those cases were included for which PCT results were available (n=184). Furthermore, as PCT levels are altered significantly in bacterial and fungal infections, cases with documented superimposed bacterial or fungal infection on microbiological culture results were also excluded (n=55). A total of 136 cases were included in the final analysis as shown in Figure 1. The demographic details alongside the length of hospital admission in the severe and non-severe groups and survivor and non-survivor groups respectively are depicted in Table 1 and Table 2. There was a total of 74 (55%) cases in the severe category and out of these 21 (28%) progressed to mortality. Whereas none of the non-severe cases i.e. 62 (45%) expired.

PCT was found to be significantly higher in the severe group compared to the non-severe cases group the median (IQR) being 0.17 (0.07-0.83) and 0.09 (0.04-0.33) ug/L respectively, *p value*= 0.01. Similarly, the PCT levels with a median value of 0.59 µg/L in the non-survivor group, were found to be significantly higher compared to survivors (*p-value* = < 0.0001). Furthermore, on a binary logistic regression model, PCT, older age and increased duration of hospital stay were revealed as variables independently associated with mortality as shown in Table 3.

ROC curve analysis was used to compare the performance of PCT as a predictor of mortality and severity. The performance of serum PCT as the only biomarker was acceptable as an AUC of 0.76 for mortality prediction and 0.62 for severity was generated respectively, taking the outcome achieved for each study participant as the reference standard as illustrated in Figure 2. A cut-off of 0.1 ug/L yielded a sensitivity and specificity of 90% and 48% for the prediction of mortality. Whereas the same cut-off had a sensitivity and specificity of 66% and 52% for severity prediction.

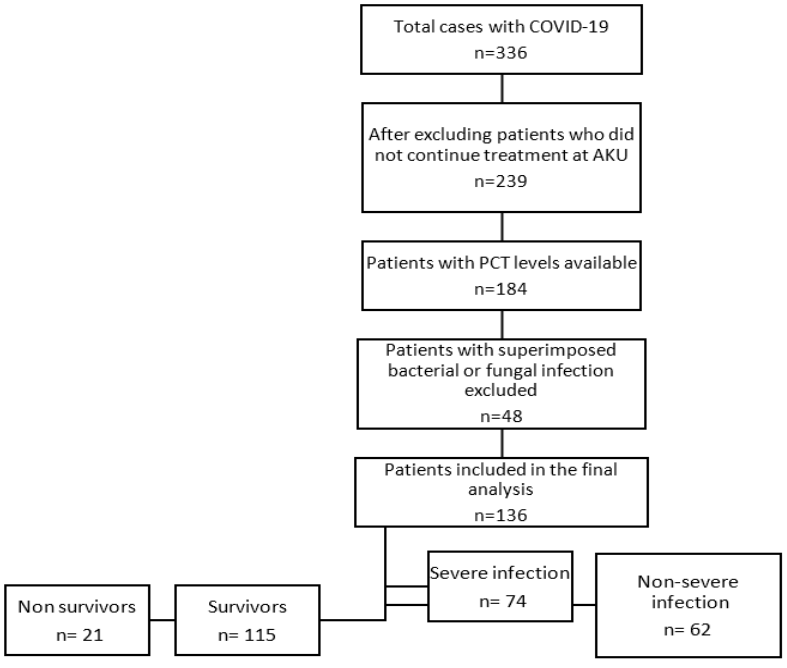

FIG. 1: Distribution of consort showing COVID-19 cases included in the final analysis.

Table 1: Demographic details and PCT in Severe Vs Non-Severe group

	Severe Cases (n=74)	Non-Severe Cases (n= 62)	*p-value*
Male: Female	56:18	45:17	0.746
Age in years (Median IQR)	57 (45-70)	51 (28.75-66.25)	0.058
Length of hospital stay in days (Median IQR)	10.5 (6-19)	6 (3-10.25)	0.000
PCT in µg/L (Median IQR)	0.17 (0.07-0.83)	0.09 (0.04-0.33)	0.015

P <0.05 statistically significant and P < 0.01 highly significant

Table 2: Demographic details and PCT in Survivor Vs Non-Survivor group

	Survivor Cases (n=115)	Non-Survivor Cases (n=21)	*p-value*
Male: Female	86:29	15:06	0.681
Age in years (Median IQR)	53 (37-66)	70 (54-78)	0.003
Length of hospital stay in days (Median IQR)	8 (4-13)	12 (3-19.5)	0.410
PCT in µg/L (Median IQR)	0.11 (0.05-0.4)	0.59 (0.17-2.6)	0.000

P <0.05 statistically significant and P < 0.01 highly significant

Table 3: Binary logistic regression model of variables independently associated with mortality

	Survivor Cases (n=115)	Non-Survivor Cases (n=21)	*p-value* (95%CI)
Male: Female	86:29	15:06	0.824 (0.341-3.859)
Age in years (Median IQR)	53 (37-66)	70 (54-78)	0.020 (0.929-0.994)
Length of hospital stay in days (Median IQR)	8 (4-13)	12 (3-19.5)	0.038 (0.910-0.997)
PCT in µg/L (Median IQR)	0.11 (0.05-0.4)	0.59 (0.17-2.6)	0.025 (0.833-0.988)

P <0.05 statistically significant

DISCUSSION

During the global COVID-19 pandemic, the role of laboratory medicine for clinical decision-making and evaluation of biomarkers for early prediction of the severity and mortality was markedly highlighted particularly for smart resource allocation in already exhausted and resource-constrained set-ups. Despite the fact much was mysterious about the rapidly evolving course of COVID-19 disease, the clinicians utilised some of the knowledge from previous experiences and knowledge of other respiratory viruses and infectious agents and utilised the prognostic biomarkers to virtuous effect. PCT whose values are established to be not substantially modified in patients with viral infections, has been widely utilised as a prognostic biomarker in the subsequent clinical course of severely ill patients with several

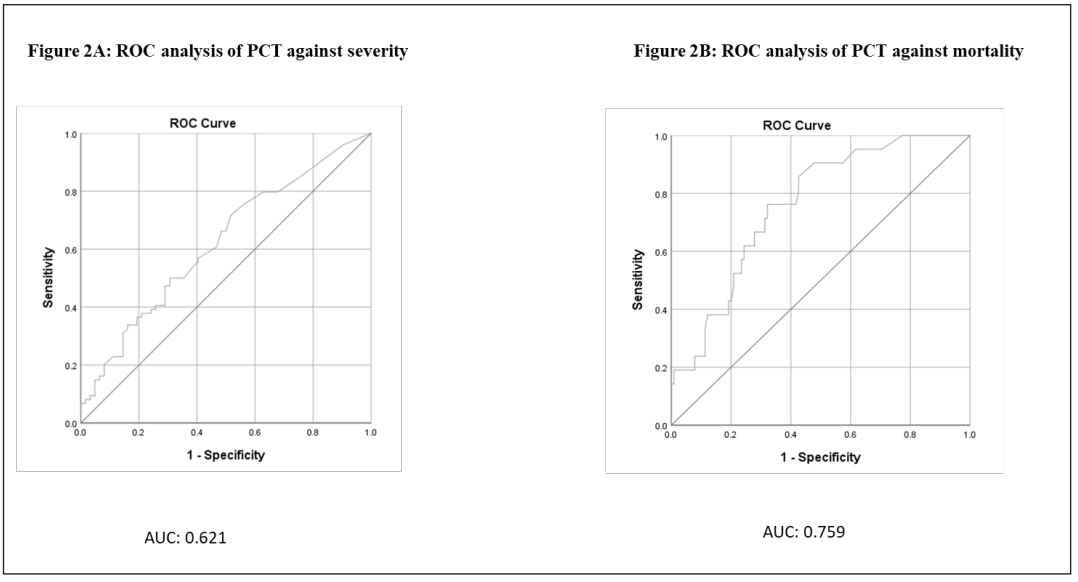

FIG. 2: ROC analysis for PCT against severity and mortality.

infectious aetiologies since its inception in 1933.[15] In context of COVID-19, the production and release into the circulation of PCT from extrathyroidal sources is thought to be massively intensified during infectious insults and furthermore, actively precipitated by the inflammatory cytokines storm and escalating serum levels of interleukin (IL)-1β, tumour necrosis factor (TNF)-α and IL-6.[7]

A meta-analysis has shown that increased PCT was associated with a fivefold higher risk of severe COVID-19 infection.[16] Furthermore, PCT value remains within reference ranges in patients with non-complicated COVID-19; any substantial increase reflects either a non-viral co-infection and the development of a severe form of disease and a more complicated clinical picture.[16] Our study reported a male predominance with COVID-19 (75%). Likewise, a retrospective cohort study of hospitalised COVID-19 cases from Lombardy, Italy reported 71% of cases being males and another report from Wuhan China reported 75%.[17,18]

Utility of PCT for the severity of infection was reflected by the statistically significant *p*-value of 0.015 and an area of 0.62 on the ROC analysis as attained by this study. Our findings are in concurrence with a prospective cohort study by Wang *et al*. in China and Ortiz-Brizuela *et al*. from Mexico, which have reported a strong association between PCT and severity of the disease progression, p-values being 0.011 and < 0.001 respectively.[19,20]

Furthermore, the increasing number of patients was an independent predictor of mortality as well as significant association was noted with severity, with the median age being 70 years and 57 years in the non-survivors and severe category. Results reported by Luo *et al*. in COVID-19 cases from China second our findings, where high proportion of mortality was noted for the age group above 60 years and increasing age was linked with disease advancement.[21] PCT emerged as a strong predictor of mortality in our cohort of patients with statistically significant results on binary logistic regression as well as an AUC of 0.76 on ROC analysis. Another study using COVID-19 from an integrated-delivery health system (Ochsner Health) in Louisiana has shown that PCT was significantly associated with in-hospital mortality.[22]

There were some limitations of this study; serial measurement of PCT were not performed to report the kinetics. Another factor to consider in our study setup was the possibility antibiotic treatment before admission, which can affect study variables, however no such details were available.

In order, to ensure better patient-centered care and resource utilisation, early identification of patients who may be at a higher risk of severe infection and mortality is pivotal. Baseline PCT concentration is a promising predictor of mortality and severity in COVID-19 cases when considered in combination with clinical details and other laboratory tests while designing the therapeutic regimen. Moreover, we propose adding PCT to the list of standard biochemical

work up of COVID-19 cases, to further improve clinical outcomes.

Authors' contribution: Sibtain Ahmed performed the literature search, data analysis and write-up of the work in the first draft. Zeeshan Ansar Ahmed, Imran Siddiqui and Naveed Haroon Rashid were involved in the laboratory workup, patient selection and critical revision of the article for the intellectual content. Maheen Mansoor performed data collection, clean up and helped with tables. Lena Jafri conceived the idea, coordinated the writing of the paper and reviewed the final draft.

Conflict of interest: The authors declared that there was no conflict of interest.

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