Original Research Article

Study of red cell distribution width as a predictor of outcomes in organophosphate compound poisoning

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Abstract

Introduction: Organophosphorus compound poisoning is a widespread problem in a developing country like India, and it is a major clinical and public health concern. There have been efforts to find novel tools/markers to assess the prognosis and the use of RDW has been proposed in OPCs poisoning, wherein RDW can be used as a predictor of outcomes in OPCs poisoning. **Objectives**: Hence our objective was to evaluate the association of RDW with the outcome of Organophosphate poisoning. **Methodology:** The study consisted of 115 patients who were admitted to JSS hospital critical care due to consumption of Organophosphorus compounds. Patients were assessed and detailed history were taken and blood investigations of Complete Hemogram and Pseudocholinesterase were sent after informed consent. **Results:** The patients were divided into 3 groups; 1) Recovered without complications; 2) Recovered but had acute complications 3) Death; with 52% patients in group A and 27% patients in group B and 20.9% were in group C. The most common complication in the group 2 was respiratory failure.

RDW as a predictor for outcomes in Organophosphate compounds has a Sensitivity of 87.5% and specificity of 51.65% with a diagnostic accuracy of 59.13%. But as an independent predictor of mortality, it was not significant. **Inference:** RDW can be used as a predictor of outcomes in Organophosphate compound poisoning cases as RDW was elevated in cases with complications and death and was found to be significant. But as an independent predictor for mortality, it was not significant.

Key words: RDW; Organophosphate Compound; Outcomes

Introduction

Organophosphorus compound poisoning is a widespread problem in a developing country like India and it is a major clinical and public health concern. Of all the acute poisoning cases presenting to the casualty, 50% of those would be usually organophosphate compounds in India[1]. These compounds are widely used in India as they are easily available in shops and are used as insecticides.

According to G. Ravi et al 2007, the annual cases of pesticide poisoning in India could be over 76 000. In addition, Gunell et al. 2007 calculated that the number of intentional suicides in India is around 126,000[2].

There have been many prognostication scores that have been developed for organophosphorus compound poisoning (OPCs) such as poisoning severity score, Peradeniya organophosphorus poisoning scale, also the use of serum Pseudocholinesterase levels (Butyrylcholinesterase), and other critical care scoring like APACHE II and GCS. Though these have all been time-tested methods that are being used to treat current OPCs poisoning cases, there have been reviews being made into this field to find a novel tool/marker to help guide the treatment.

Red Cell Distribution Width in the recent past has been used as an independent predictor of prognosis among patients with cardiac conditions like coronary heart disease, such as acute myocardial infarction, congestive heart failure, and coronary heart disease. It has also been used as predictors in conditions like stroke, community-acquired pneumonia, septic shock, and acute pancreatitis[3,4,5].

Red cell distribution width (RDW) is routinely assessed as part of the complete blood count (CBC) to gather information on the heterogeneity in the size of circulating erythrocytes. Computationally, RDW is the coefficient of variation of the mean corpuscular volume (MCV) and therefore higher RDW values reflect greater heterogeneity in MCV (anisocytosis), which is usually caused by perturbation in erythrocyte maturation or degradation.

In a much simple sense; Red Blood Cell distribution width (RDW) is a simple measure of the broadness of erythrocyte size distribution, conventionally called anisocytosis. This measure is easily, inexpensively, and rapidly calculated as a ratio of standard deviation (SD) of red blood cell (RBC) volume and mean corpuscular volume (MCV) [i.e., (RDW-SD)/(MCV)×100], with the result expressed as a percentage.Given that the RDW is routinely reported by clinical laboratories as a component of the CBC and is available for most patients, understanding its prognostic significance could be very valuable for risk stratification in clinical decision-making.

A similar application of the RDW has been proposed in OPCs poisoning, where RDW can be used as a predictor of outcomes in OPCs poisoning. There have been several attempts to find out whether RDW can in fact be used as a predictor of outcomes, but most of these studies so far have been done on a retrospective basis.

Organophosphorus (OP) compounds and carbamates, also known as cholinesterase inhibitors, are widely used pesticides. These agents,

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which comprise thousands of structurally related substances, are responsible for a large number of suicidal or accidental poisonings, with the greatest mortality (an estimated 200,000 deaths per year) in rural areas of developing countries.

A great variety of substituents is possible with R1 and R2 may be alkyl, alkoxy, aryloxy, amido, mercaptan, or other groups. With the X typically a conjugate base of a weak acid, a halide, cyanide, a thiocyanate, phenoxy, Thiophenoxy, phosphate, thiocholine, or carboxylate groups.

Organophosphorus compounds inhibit two enzymes: acetylcholinesterase (AChE), found in synaptic junctions and in red blood cells (RBCs), and butyrylcholinesterase, also known as pseudocholinesterase (PChE) or plasma cholinesterase, found in the blood. Each of these enzymes breaks down acetylcholine.

Blockade of AChE is the most clinically significant effect of OPs and carbamates because this leads to the accumulation of excessive amounts of acetylcholine at muscarinic receptors (found on various cholinergic secretory cells), at nicotinic receptors (located on skeletal neuromuscular junctions and autonomic ganglia), and in the CNS.

Normally the cholinesterase rapidly hydrolyzes the neurotransmitter acetylcholine into inactive fragments of choline and acetic acid after the completion of the neurochemical transmission. The major toxicity of organophosphate compounds is the covalent binding of phosphate radicals to the active sites of the cholinesterase, transforming them into enzymatically inert proteins. Organophosphates thus act as "irreversible cholinesterase inhibitors" because the organophosphate-cholinesterase bond is not spontaneously reversible without pharmacological intervention. The inhibition of cholinesterase activity leads to the accumulation of acetylcholine at synapses, causing overstimulation and subsequent disruption of transmission in both the central and peripheral nervous systems. Exposure to organophosphate compounds will, therefore, interfere with synaptic transmission peripherally at muscarinicneuroeffector junctions and nicotinic receptors within sympathetic ganglia and at skeletal myoneural junctions. This is accomplished by overstimulation of acetylcholine receptor sites that lead to a variety of physiologic and metabolic derangements. Disruption of transmission also will occur at the acetylcholine receptor sites within the central nervous system.

Clinical manifestations

The clinical manifestation of OP poisoning depends on the agent, quantity, and route of entry. Ingestion and inhalation result in the more rapid development of symptoms than dermal exposure. After ingestion symptoms appear within 30-90 minutes and a maximum of 24 hrs. in the case of compounds that are highly lipophilic, and which require metabolic bioactivation.

Local Effects

GI symptoms appear first before the onset of systemic symptoms. Inhalation typically exhibits respiratory effects. After ocular exposure symptoms generally begin in the eyes.

Systemic Effects

Three well defined clinical phases are observed

1) Initial cholinergic phase.
2) The intermediate syndrome (IMS)
3) Delayed polyneuropathy

The cholinergic phase

It is mainly due to the accumulation of Ach at the cholinergic synapses and may be classified into

1) Muscarinic (all postganglionic nerve endings)
2) Nicotinic (Autonomic ganglia and skeletal muscle endplates).
3) CNS manifestations (synapses in CNS)

Commons signs and symptoms of acute toxicity are

Dumbels: Diarrhea, Urination, Miosis, Bronchospasm, Emesis, Lacrimation, Salivation

Symptoms may occur within 5 minutes following large ingestion and death is possible within 15 minutes. Most patients will be symptomatic within 8 hours and almost all patients within 24 hours if consumed.

Objectives of the study

Primary objective

"To evaluate the association of RDW with the outcome of Organophosphate poisoning"

Secondary Objective

"To assess the association between serum pseudocholinesterase levels and RDW"

Methodology

Study Design

Cross-sectional observational study conducted in the Hospital

Study Duration

18 months -November 1st, 2019 to April 30th, 2021

Sampling technique

Random sampling; Taking previous study of Chang-Woo et al as reference sample size was calculated to be 101.

Study setting and Method of collection of data

This study was cross-sectional, observational and was conducted on patients admitted to the General medicine Department who fulfilled the inclusion criteria which is clinical assessment for symptoms associated with OP compound consumption (Bronchospasm, bradycardia Salivation, lacrimation, urination, defecation, gastric emesis, and bronchorrhea) and history of consumption. Exclusion criteria have been listed and patients under this will not be considered for the study

b. Exclusion Criteria

1. Patients <16 years of age
2. Co ingestion with other agents
3. Cardiac arrest prior to hospitalization
4. Transfer out to a different hospital
5. Patients going DAMA
6. Patients with previous Hematological disorders
7. Patients with other active infections
8. Patients with malignancies
9. Patients with Anemia

Study Conduct

Clinical study

A total of 126 cases presented to JSS Hospital during the duration of the study; 115 were taken as participants in the study, 9 cases were excluded as they did not fulfill the inclusion criteria or were part of the exclusion criteria.

Following parameters were collected from the patient after an informed consent

Demographic data, Initial vital signs, Levels of consciousness (AVPU scale), initial laboratory measures were obtained upon admission, Pseudo Cholinesterase.

The Hematological components like RDW were all done by the SYSMEX XN 1000 hematological analyzer.

Blood urea nitrogen, Creatinine, and Albumin levels were all done by autoanalyzer, Roche-Cobas 6000 with these following methods

BUN (Blood urea nitrogen) - Urease UV method

Creatinine – Enzymatic IFCC/ IDMS method

Albumin – Bromocresol green

Pseudocholinesterase- Butyrylthiocholine

Patients were placed into three Categories

1) Recovered with no complications -GROUP A
2) Recovery with Acute complications – GROUP B
3) Death – GROUP C

After the data was compiled, the data analysis was done by SPSS software version 21. The descriptive variables like Mean, standard deviation, Frequency, and Percentage of all the above were done.

The inferential variables were calculated using the Chi-square test and Spearman's Correlation was done. ANOVA will be used to compare between the groups.

Regression analysis was done Pie charts, Error bars and scatter plots and ROC curves were plotted with Cut-offs for RDW.

Results

Table 1: Demography

		Count	%
Age	<25	21	18.3%
	26-40	60	52.2%
	41-55	20	17.4%
	>55	14	12.2%
Sex	Female	22	19.1%
	Male	93	80.9%
CO-morbidities	No	75	65.2%
	Yes	40	34.8%

In our Study had most of the cases -52.2% cases- were seen between 26 – 40 years of age group, followed by 18.3% cases aged less than or equal to 25, 17.4% cases in the age between 41 – 55 years of age and 12.2% cases had age more than 55.

80.9% male cases were observed in this study where 19.1% were female cases.

It was also noted that comorbidities were seen in 34.8% cases where as 65.2% cases were presented without any comorbidities.

On arrival to the hospital 76.5% cases were seen having normal blood pressure where 13.0% cases presented with hypotension and 10.4% cases were observed to have hypertension.

76.5% cases were seen with low severity, 17.4% cases were seen having severe severity and 6.1% cases were seen with moderate severity.

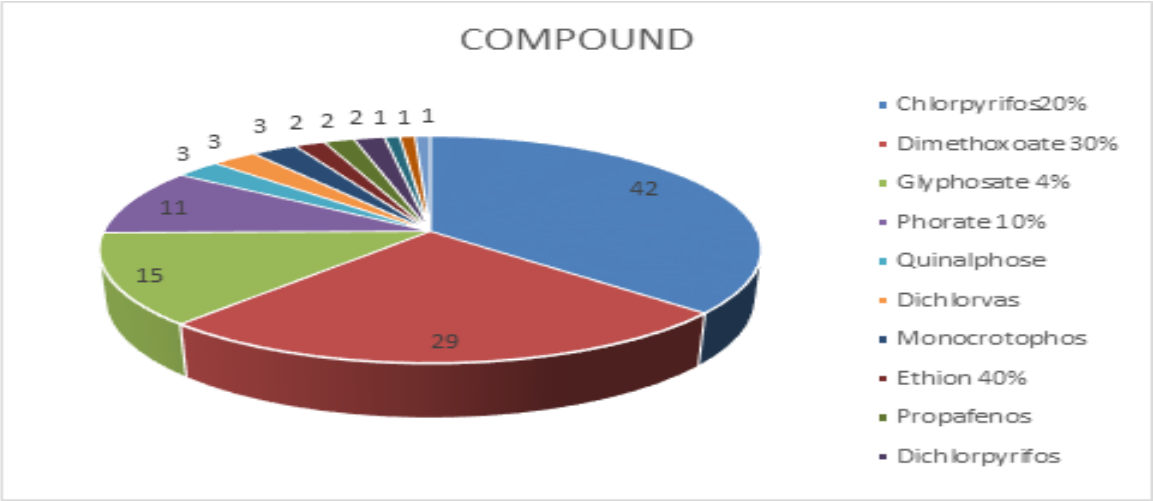

Figure 1: Showing Different Compounds

Chlorpyrifos20% was the most commonly used compound followed by dimethoxoate30%

The patients belonging to group B and group C (Recovered from acute complications and death); had the following complications, 19.1% cases were presented with respiratory failure, 5.2% cases had intermediate syndrome, 3.5% cases were seen having severe Cholinergic crisis and Atropine delirium while 2.6% had respiratory failure + HIE respectively, 1.7% each cases were seen having Respiratory failure + CNS manifestations, Respiratory failure + hypotension, Respiratory failure + severe acidosis and ventricular tachycardia respectively, 0.9% of each were seen having Aspiration pneumonia, Atropine, Atropine delirium and acidosis, Hypotension, Intermediate syndrome + Seizures, severe acidosis, toxic myocarditis, UGI bleed, Urethral injury and ventilator ass pneumonia respectively.

While 48.7% of the patients did not develop any complications and were placed in group in group A.

Table 2 Outcomes Of Study

OUTCOMES	Count	%
GROUP A (Recovered without complications)	60	52.2%
GROUP B (Recovered with complications)	31	27.0%
GROUP C (Death)	24	20.9%
Total	115	100.0%

The outcomes that have been previously outlined had the following distribution 52.2% cases were Recovered without complications, 27.0% cases were Recovered with complications and whereas 20.9% mortality was seen.

Table 3: ANOVA analysis with respect to outcomes

	Outcomes								
	Group A (Recovered without complications)		Group B (Recovered with complications)		Group C (Death)		Total		
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	p

Age	33.4	10.5	34.5	12.7	48.0	16.7	36.7	13.8	<0.0001
Saturation	95.77	4.91	86.29	14.25	83.25	12.64	90.60	11.34	<0.0001
Respiratory rate	21.05	3.60	25.58	5.97	26.96	4.97	23.50	5.28	<0.0001
Pulse rate	101.45	19.40	109.03	18.12	112.83	22.22	105.87	20.10	0.04
RDW	13.33	.95	13.50	1.27	13.97	.84	13.51	1.05	0.04
Cholinesterase	2756.08	3343.56	1544.87	2445.09	1449.83	2769.81	2162.34	3054.35	0.1

With respect to outcome of the present study, all the above parameters were tested by ANOVA and has been shown in the table above.

13.33 mean RDW was seen in group A, 13.50 mean RDW was seen in group B cases and 13.97 mean RDW was seen in group C cases. Statistical significance was seen between these groups proving that there is an association between RDW and outcomes of OP compound poisoning patients.

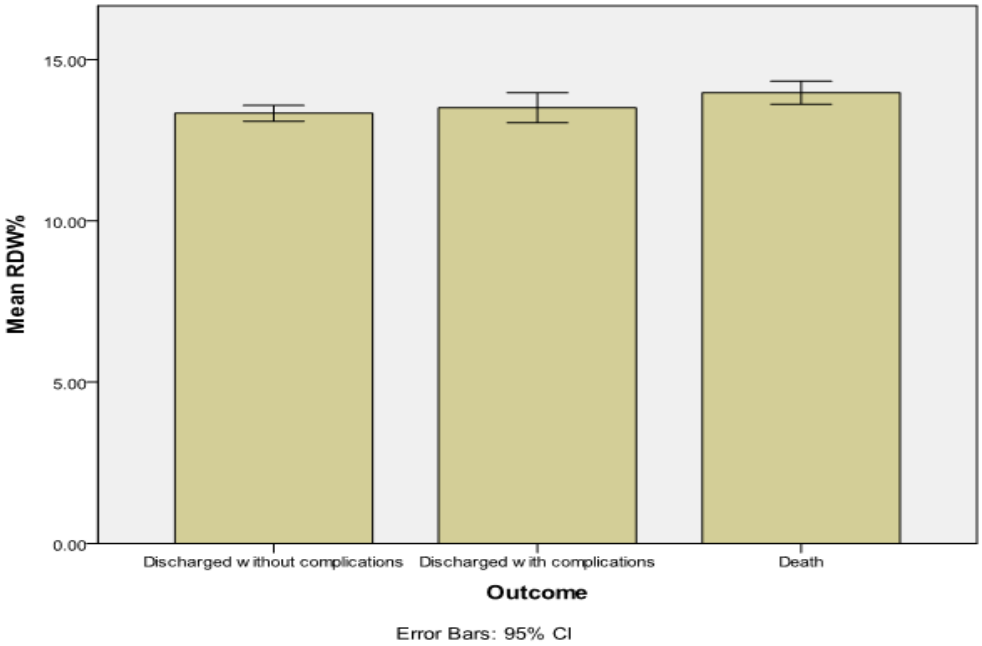

Figure 2: Error bar depicting co-relation with RDW

In this study, the Pseudo Cholinesterase levels were grossly reduced in group C; that is patients who succumbed and moderately low in group B where patients who had acute complications and were discharged, while higher levels of Pseudo cholinesterase were seen in group A. This was also found to be statistically significant (p = 0.04 by Kruskal Wallis test). As it can be seen, illustrated here in the above table and graph Pseudo Cholinesterase levels can predict the outcome in OP compound poisoning.

Table 4: Regression analysis of variables with respect to mortality

							95% C.I.for OR	
	B	**S.E.**	**Wald**	**df**	**p**	**OR**	**Lower**	**Upper**
Age	.098	.041	5.618	1	.018	1.103	1.017	1.195
Co-morbidity	-1.022	1.200	.726	1	.394	.360	.034	3.778
Blood pressure			2.462	2	.292			
Hypotension	2.259	1.440	2.462	1	.117	9.573	.570	160.906
Hypertension	.559	1.364	.168	1	.682	1.750	.121	25.325
Saturation	.082	.063	1.703	1	.192	1.086	.959	1.229
Respiratory rate	.175	.142	1.508	1	.219	1.191	.901	1.575
Pulse rate	.012	.028	.181	1	.671	1.012	.959	1.068
Conscious			6.842	3	.077			
VERBAL	.267	1.522	.031	1	.861	1.306	.066	25.781
PAIN	4.554	2.036	5.005	1	.025	95.005	1.758	5134.255
UNRESPONSIVE	3.005	1.318	5.195	1	.023	20.182	1.523	267.388
RDW	.970	.545	3.168	1	.075	2.639	.906	7.682
Cholinesterase	.000	.000	.147	1	.702	1.000	.999	1.000
Constant	-30.3	14.716	4.232	1	.040	.000		

In our present study, RDW was not found to be significant as an independent predictor of mortality in patients with OP compound poisoning. As the Table above shows, it was found to be statistically not significant.

Pseudo Cholinesterase and RDW both as independent predictors were able to predict the outcomes in patients with OP compound poisoning. But no association could be found between the two.

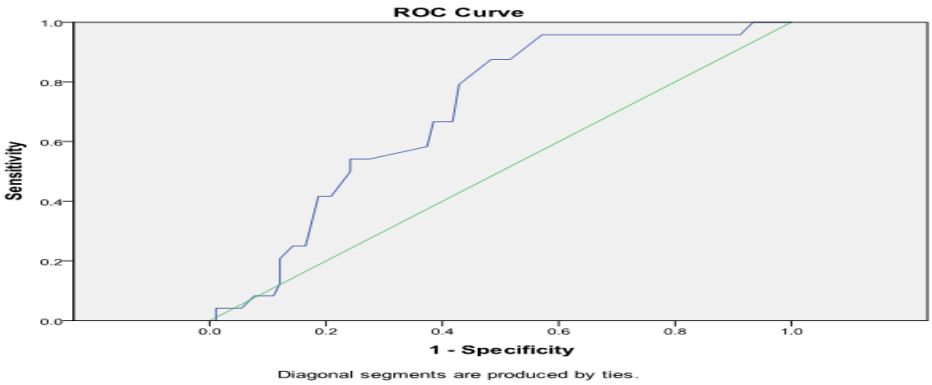

Figure 3: ROC curve for RDW with respect to outcomes

In present study cases who had RDW count more than 13.25, 32.33% mortality was seen where 67.7% cases were discharged either with complications or without complications. Cases who had RDW less than 13.25, 6% mortality was seen, and 94% cases were discharged with or without any complications. (p value 0.001

87.5% Sensitivity, 54.65% specificity and negative predictive value of 94% was seen with respect to RDW and outcome of cases. It has a 59.13% diagnostic accuracy to be used for prognosis of patients admitted with OP compound poisoning and their duration of stay.

Discussion

Organophosphorus compound poisoning is a commonly encountered problem in India, due to limitations of resources, a cheaper diagnostic test would be a welcome change. In this present study, we have tried to find such an alternative in the form of RDW.

RDW which is a measure of anisocytosis is commonly measured by automated hematology analyzers, which has been used in the differentiation of various anemias. It has been shown that RDW is associated with poor prognosis in various vascular disorders and sepsis and other acute conditions[6-14].

The mechanisms behind this have not been clearly defined but, it is thought that systemic inflammation and oxidative stress increase the RDW[15-18]. Suppression of maturation of the RBCs in bone marrow by the inflammatory cytokines led to immature RBCs entering the circulation and in effect increasing the RDW. RDW elevation has been closely linked with Oxidative stress[19], anisocytosis is increased by disrupted erythropoiesis secondary to oxidative stress, increased oxidative stress contributes to more lipid peroxidation and a reduction in the phospholipid composition of the RBC membrane. Which results in the erythrocyte membrane being damaged and erythrocytes losing their integrity[20]. The consequence being, the erythrocyte membrane being deformed, as a result, the lifespan of erythrocytes is shortened[21].

it has also been found to destroy and shorten the survival time of RBCs. Organophosphorus poisoning has been known to set of inflammatory processes with increased production of inflammatory cytokines like Interleukin 1β and IL- 8.25 Although the mechanisms of how RDW is influenced by OP poisoning, it has been shown to be associated.

Elhosary NM et al (2018)[22] in their study they observed cases aged between 18 and 60 years who were poisoned with different types of pesticides. Acikalin A et al (2017)[23] observed the mean age was 32.4±15.0 ranging from 13 – 94 yrs. In their study, 61.2% were the female case and 38.8% were male cases. Dundar ZD (2015)[24] observed the median age of the patients was 37.0 (25.5-50.8) years. They also observed 40 (55.6%) patients were male cases and 32 (44.4%) were female cases. Kang C et al (2014)[25] in their study 57.5 yrs. mean age was observed. Out of 102 cases, 68 (66.7%) were male cases and 34 (33.3%) were female cases.

Elhosary NM et al (2018) in their study observed 87.5% of cases had consumed accidentally, 12.5% cases consumed in a suicidal manner, 25% cases of ingestion. They observed a higher percentage of patients exposed to the anticholinesterase's insecticides including organophosphates and carbamates through mixed routes as inhalation, skin, and or ingestion. Acikalin A et al (2017) observed that patients with suicidal intentions had oral intake which was 60 patients (75%), exposure while farming for in the field in 11 patients (13.8%), accidentally consumed along with food orally was seen in five patients (6.2%), and wiping on the head as a treatment for pediculosis in four patients (5%).Dundar ZD (2015) et al in their study they observed the route of exposure was oral ingestion in 56 (77.8%) patients, inhalation in 12 (16.7%) patients, and transdermal in 4 (5.6%) patients.

In the present study, 76.5% of cases were seen having normal blood pressure where 13.0% cases were presented with hypotension on arrival and 10.4% cases with hypertension on arrival. Which also had an effect on the prognosis which will be described later.

Kang C et al (2014) in their study out of 149 cases 24 were presented with Unresponsive on the AVPU scale.

In the present study while presenting in hospital 53.9% of cases were awake, 21.7% cases were unresponsive, 17.4% cases were verbal, and 7.0% cases were presented with pain.

It was observed that around 76.5% cases were seen with OP compounds with Low toxic type, 17.4% cases were seen having Highly toxic compounds, and 6.1% cases were seen with moderate toxicity compounds.

Complications that were commonly seen in the patients belonging to group B and group C (Recovered from acute complications and death), were listed above it was observed that respiratory failure and cholinergic crisis were the most common symptoms that were observed. There were other acute complications that were rarely seen.

As discussed earlier the outcomes in the study were divided into 3 groups. 52.2% of cases were discharged without any complications, 27.0% of cases were patients who were discharged after recovering from acute complications that prolonged their hospital stay and a mortality of 20.9% was seen.

Mortality was more commonly observed in the older age group as explained above. With the patients who died the mean age was around 48 years old. Younger patients have a better prognosis when compared to older patients.

RDW was seen to be highest in mortality cases and mildly deranged in patients who were discharged after recovering from acute complications. While it was lower in patients who were discharged

without any complications. Hence it can be used as a prognostication score for patients.

Serum pseudocholinesterase levels were measured for all patients and it was seen that it also can be used as a prognostication test as we know. In our study, there was evidence for the same. ANOVA tests were not significant

Serum pseudocholinesterase levels were measured for all patients and it was seen that it also can be used as a prognostication test as we know. In our study, there was evidence for the same (Kruskal-Wallis test as values were largely staggered).

Elhosary NM et al (2018) observed hematological parameters, patients who died had significantly higher RDW% (P <0.001). From the ROC curves, patients were considered of bad prognosis when RDW% were ≥14.3 (p-value <0.001) which is comparable with our study.

Patients presenting with hypotension were found to succumb to the effects of the poison and also were found to develop complications than patients with normal blood pressure; it was also noticed that hypertensive presentations of patients also indicated a poorer prognosis with patients either developing complications or succumbing to the poison.

Elhosary NM et al (2018) observed most sensitive hematological parameter was RDW (93.8%). Dundar ZD et al (2015) RDW was higher in non-survivors than survivors [15.40 (15.10-16.40) and 14.30 (13.30-16.00), respectively, p=0.047). Also, 14.35 mean RDW% was observed in all the cases. Kang C et al (2014) in their study Patients with an RDW more than 13.5% were more on the older age of the spectrum when compared to the other group. The mortality rate within 30 days after admission due to Organophosphorus compound poisoning was 20.6% (21 of 102). Patients who succumbed had significantly higher levels of RDW (13.9±1.8 vs 12.9±0.8, P=.013; and older (72.2±9.1 vs 53.7 ± 14.7, P b .001). The cut-off point for RDW was more than 13.5%, and the area under the ROC curve was 0.675 (95% CI, 0.522-0.829, P=.013). In multivariate analysis, age, unresponsive in AVPU scale, and RDW more than 13.5% were independent prognostic factors for 30-day mortality. Patients with an RDW of more than 13.5% had a 2.64-fold increased risk of 30-day mortality than did patients with an RDW less than or equal to 13.5% during the follow-up period

In the present study cases who had RDW count more than 13.25, 32.33% mortality was seen where 67.7% cases were discharged either with complications or without complications. In cases who had RDW less than 13.25, 6% mortality was seen, and 94% cases were discharged with or without any complications.

Dundar et al observed RDW% 73% sensitivity and 70% specificity. Similarly, Kang et al showed that when the RDW was more than 13.5%, the sensitivity was 57.1%, and the specificity was 84.0% (positive predictive value, 48.0%; negative predictive value, 88.3%) which is comparable with our study.

It was found that RDW as a predictor of outcomes had 87.5% Sensitivity, 54.65%, and a negative predictive value of 94%. This shows that there is indeed an association between RDW and Outcomes of OP compound poisoning.

RDW as an independent predictor for mortality was not seen in our study, with regression analysis showing that it was not significant when compared to all the previous studies that we have shed light on, this is a deviation from the previous studies where they showed that RDW could be used as an independent predictor for Mortality.

Our study also checked the association between RDW and Serum pseudocholinesterase levels, if they co-related with each other as one has been used in standard practice in OP poisoning and the other is being considered for the same, but there was no association found between the two.

Limitations

Our present study was done as a cross-sectional study against all the previous studies which were mainly retrospective lacks a larger sample size and was mainly based on one institution. So, any further investigations into the same topic require a larger sample size based out of multiple institutions with more varied demography.

RDW was not found to be independent predictor of mortality in our study, which is a deviation from other studies done by Kang et al; Dundar et al which has to be revisited in a larger, multicentric trial.

Conclusion

We would like to conclude that, RDW is associated with outcomes in Organophosphate compound poisoning. Measuring RDW can help us prognosticate the outcome of patients with organophosphate compound poisoning. Hence, we think that this can be used as a cheaper tool to prognosticate OP compound poisoning patients.

It is to be noted though that as per our study it cannot be used as an independent predictor for mortality in these patients.

It also can be used to predict the duration of hospital stay in these patients as mentioned above.

For our secondary aim, though pseudo cholinesterase levels and RDW could individually be predictors of outcomes in OP compound poisoning, there was no association that could be found between the two.

Conflict of interest

No conflict of interest

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