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RESEARCH ARTICLE

ANTIPLATELET DRUG ACTIVITY EVALUATION IN PATIENTS OF ACUTE CORONARY SYNDROME IN CENTRAL INDIA: IN THE SCENARIO OF ANTIPLATELET DRUG RESISTANCE

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ABSTRACT

Background: Antiplatelet drugs are the cornerstone in the management of acute coronary syndrome. Recent studies suggest variability in response to aspirin and clopidogrel. A few patients on preventive therapy with aspirin and/or clopidogrel got recurrent attacks of thromboembolic episodes, further pointing towards an uncertain response. **Method:** A cross sectional, observational study was conducted in a tertiary care rural hospital in central India. 102 patients with a diagnosis of acute coronary syndrome who were prescribed aspirin and or clopidogrel for at least 7 days as antiplatelet therapy were included in the study. The samples were tested for platelet function by a test which was an adaptation of plateletworks kit. **Results:** On evaluation, the prevalence of aspirin resistance was 45.09%, of which 21 % were non responders while 24% were semi responders. Clopidogrel resistance was found to be 76.13%, of which 29% were non responders while 47% were semi responders. **Conclusion:** Our study shows an upsurge in aspirin and clopidogrel resistance. Standard yet less expensive, affordable, specific and sensitive platelet function assays are needed to see the antiplatelet aggregation activity to assure the drugs activity to the fullest.

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INTRODUCTION

Antiplatelet drugs are the cornerstone in the management of acute coronary syndrome (ACS) which is a thromboembolic phenomenon. It encompasses acute myocardial infarction (AMI) with or without ST segment elevation and unstable angina (Huffman, 2009). Aspirin alone or in combination with clopidogrel is used for secondary prevention of thromboembolic episodes (Maree, 2007). Clinical trials have shown the efficacy of aspirin as well as of clopidogrel in secondary prevention of cardiovascular disease (Maree , 2007; Antiplatelet Trialists', 1994 & 2002). Furthermore, complementary mechanisms of action of aspirin and clopidogrel translate into additive benefit in certain populations. Recent studies have suggested a variable platelet response to aspirin and clopidogrel in atherothrombotic diseases (Maree, 2007). Additionally, few patients already receiving preventive therapy in terms of aspirin and/or clopidogrel got recurrent attacks of thrombo-embolic episodes (Antithrombotic Triatlists, 2002), further pointing towards variable responses. Studies suggesting emergence of aspirin and clopidogrel resistance (sole or dual) have strengthened the above findings and posed another therapeutic challenge in cardiovascular medicine (Wang, 2006; Guyer, 2009; Matetzky, 2004). Startled by these studies, in India too, the resistance and variable response to antiplatelet drug therapy has been explored by a few researchers (Sadiq, 2005; Kumar, 2007; Thomson, 2009; Guha, 2009 and Guha, 2009). Variable response to aspirin and clopidogrel has resulted in recurrent thromboembolic episodes even in patients already on these drugs causing significant morbidity and mortality. The term "resistance" used in this context denotes the inadequate response or treatment failure because of various reasons like improper drug compliance, early discontinuation, possible drug interactions, inadequate dose, increased platelet turnover, genetic polymorphisms, potential bypass mechanisms and others (Guyer, 2004). Some use it to refer to the continued occurrence of ischemic events despite adequate anti-platelet therapy and compliance. With availability of objective tests, the term is still evolving. The global prevalence of resistance to aspirin is 5.5% to 60% (Gasparyan, 2008) and to clopidogrel 16.8% to 21% (Snoep, 2007). In India, incidence of aspirin resistance was documented at 38.1% by Thomson et al (Thomson, 2009). In a study by Guha et al (2009) aspirin, clopidogrel and dual drug resistance were encountered in 35%, 72.5% and 32.5% patients with recurrent ACS, respectively while the corresponding figures for patients with first episodes of ACS were 25.3%, 42.3% and 18.8% respectively. The studies available in India are few and needs further exploration. Hence, a study was planned to evaluate the level of antiplatelet aggregation activity among the patients of acute coronary syndromes who were receiving aspirin or clopidogrel alone or in combination and to assess the prevalence of resistance to these drugs in our setting.

MATERIAL AND METHODS

Ours was a cross sectional, observational study conducted in a tertiary care rural hospital in central India. Patients attending medicine OPD or admitted to medicine wards with a diagnosis of acute coronary syndrome or follow up cases of acute coronary syndrome, who were prescribed aspirin and or clopidogrel for at least 7 days as antiplatelet therapy, were included in the study after obtaining their informed written consent and explaining the study objectives. An ethical clearance from the Institutional Ethics Committee was obtained prior to the commencement of the study. Inclusion criteria: Patients with a diagnosis of acute coronary syndrome including ST elevated acute myocardial infarction (STEMI), Non-ST elevated acute myocardial infarction (NSTEMI) and unstable angina and follow up cases of the above diagnosis attending medicine OPD or getting admitted to medicine ward were included in the study.

Exclusion criteria: (a) Concurrent use of non-steroidal anti-inflammatory drugs. (b) Family or personal history of bleeding disorders. (c) Platelet count < 150 x 10^3 /L or > 450 x 10^3 /L. (d) Consent not given for participation in the study.

Around 110 patients were screened for inclusion in the study, out of which 8 patients were excluded as 3 of them had platelet counts less than 150 x 10^3 while 5 patients refused to give consent for blood sample collection. Thus, at the end 102 patients were included in the study. After filling the questionnaire regarding demographic details of the patient, medical history, medication history and biochemical details, blood sample of 4 ml was taken for platelet function assay from antecubital vein using a 21 gauge needle. The blood sample was distributed in following test tubes (a) EDTA tube- (2 ml blood sample), (b) 0.9 ml each in 2 test tubes containing 100ul of sodium citrate as anticoagulant.

These samples were tested for platelet function by a test which was an adaptation of plateletworks (Lau, 2002; Gerhard Vogel, 2008 and Sackett, 2000) kit. Plateletworks kit has agonist added in the test tube along with sodium citrate as anticoagulant and buffers. So, when the blood sample is added to it, within 5 minutes it has to be run on a coulter machine to measure the platelet count. So, portable coulter machine is needed for counting or the sampling has to be done in/near the lab. Thus, this test has a disadvantage of limited time of testing after which results are not valid.
In our study we collected sample in test tubes containing only sodium citrate and buffers. So, we had the time for transporting the sample to the lab after which we could process the sample by adding agonists. Thus, our test required only 4 ml of blood sample, had time for transportation and was convenient. In the pathology laboratory, EDTA tube was first run in the impedance based coulter machine and a baseline platelet count was noted. ADP 10uM solution was added to one of the test tube containing sodium citrated blood and after 5 minutes sharp, the tube was run on the same cell coulter machine and platelet count was noted. Similarly, collagen 2ug/ml was added to other test tube containing sodium citrated blood and after 5 minutes, it was also run to note down the platelet count. The agonists (ADP and collagen) stimulate only those platelets which were functional (not inhibited by the drug being tested) to aggregate into clumps which are not counted as platelets by the cell coulter machine, rather they got counted as lymphocytes. The difference in platelet counts provided a direct measurement of platelet aggregation and was reported as percent aggregation as per the following equation:

Baseline platelet count – Agonist platelet count x 100 = % Aggregation

Baseline platelet count

The addition of ADP 10 µM as agonist tested inhibition of platelet aggregation activity of clopidogrel while collagen 2 µg/ml as agonist tested the inhibition of platelet aggregation activity of aspirin.

Definition of low response: Patient with ≤50% collagen (2µg/ml) were labeled as aspirin responder, ≥ 75% aggregation with collagen (2µg/ml) will be labeled as aspirin non-responder, whereas 75-50 % aggregation with collagen (2µg/ml) were termed aspirin semi-responder. Thus, Patients having ≥50% aggregations with collagen (2µg/ml) were aspirin resistant (non-responders & semi-responders). Patients with ≤50% aggregation with ADP (10µM) were labeled as clopidogrel responder; 50-75 % aggregations were labeled as semi-responders and ≥75 % were labeled as non-responders. Thus patients having ≥50% aggregations with ADP (10µM) were termed clopidogrel resistant (non-responder plus semi-responder). Dual resistance was defined as ≥50% aggregation with both collagen (2µg/ml) and ADP (10µM).

Comparison of test used in the study with standard: The assay test used in the study based on plateletworks kit was compared with platelet aggregometer (standard). Platelet function assay of 10 patients who received antiplatelet drugs was measured by both the test and correlation was measured. The test used in the study was found to have a good correlation with the gold standard which was platelet aggregometer.

Statistical analysis: Statistical analysis was done by using descriptive & inferential statistics using chi square test and z-test. The statistical softwares used in the analysis were graph pad prism 5.0 and SPSS 17.0. The level of significance was 5%.

RESULTS

102 patients of acute coronary syndrome were studied of which 102 received aspirin and 88 received clopidogrel.

Table showing correlation of ADP induced aggregation by the two tests

	Mean	Std. Deviation	n	Pearson's Correlation	Spearman's Rank Correlation
Study test	55.50	24.16	10	0.94 P=0.000 S, p<0.05	0.96 P=0.000 S, p<0.05
Standard	52.90	20.13	10		

Line of Regression = -4.23+ 1.12* X, R=0.94

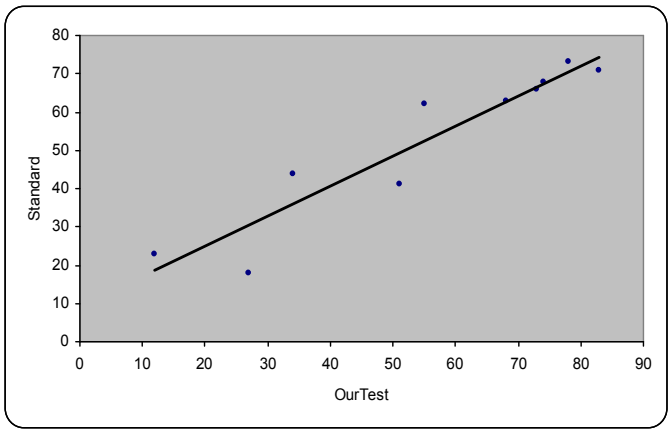

Table showing correlation of Collagen induced aggregation by the two tests

	Mean	Std. Deviation	n	Pearson's Correlation	Spearman's Rank Correlation
Study test	48.10	19.55	10	0.92 P=0.000 S, p<0.05	0.90 P=0.000 S, p<0.05
Standard	45.30	20.41	10		

Line of Regression = 7.99+ 0.88* X, R=0.92

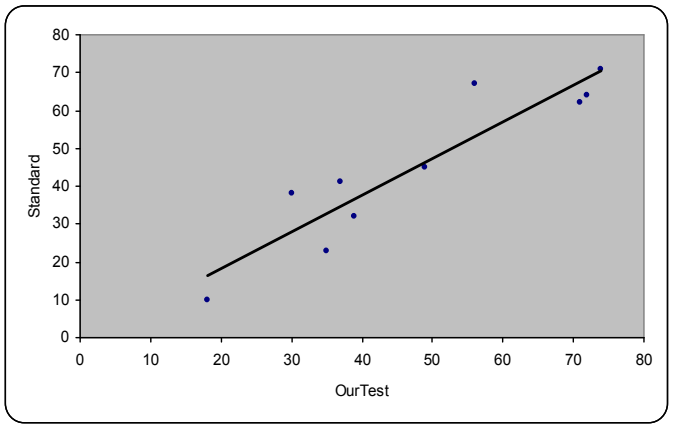

Table 1. Comparison of demographic characteristics of aspirin response groups

Variables	Aspirin R (n=56)	Aspirin SR + NR (n=46)	p-value
Age (years)	59.67 ± 10.74	60.06 ± 11.50	0.862 NS, p>0.05
Gender (M:F)	37:19	26:20	0.32 NS, p>0.05
BMI (Kg/m$^{2)}$	27.09 ± 3.61	26.90 ± 3.76	0.806 NS, p>0.05

R- Responder, SR- Semiresponder, NR- Nonresponder, NS- not significant

Table 2. Comparison of demographic characteristics of clopidogrel response groups

Variables	Clopidogrel R (n=21)	Clopidogrel SR + NR (n=67)	p-value
Age (years)	59.90 ± 11.16	59.01 ± 11.12	0.75 NS, p>0.05
Gender (M:F)	11:10	43:24	0.33 NS, p>0.05
BMI (kg/m^2)	27.63 ± 3.17	26.95 ± 3.71	0.41 NS, p>0.05

R- Responder, SR- Semiresponder, NR- Nonresponder, S- significant, NS- not significant

After evaluating the antiplatelet aggregation activity of the aspirin and clopidogrel in patients of acute coronary syndrome by a test which was adaptation of plateletworks kit, we found that the mean antiplatelet aggregation activity was 50.95 ± 24.02 and 37.28 ± 27.28 in patients on 75 mg and 150 mg of aspirin, respectively (Figure 1). The mean antiplatelet aggregation activity was 46.88 ± 23.89 in patients receiving 75 mg of clopidogrel (Figure 2). After evaluation, we found that the prevalence of aspirin resistance was 45.09%, of which 21 % were non responder while 24% were semi responder. (Figure 3) Clopidogrel resistance was found to be 76.13%, of which 29% were non responder while 47% were semi responders. (Figure 4) Dual resistance was seen in 40% of those who were prescribed both aspirin and clopidogrel. (Figure 5).

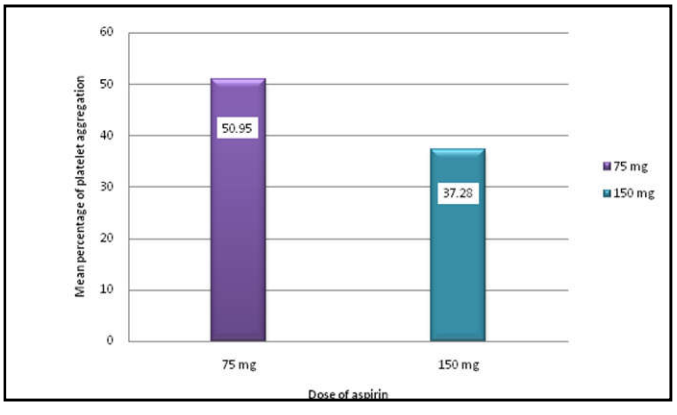

Figure 1. Antiplatelet aggregation activity of aspirin by different doses of aspirin

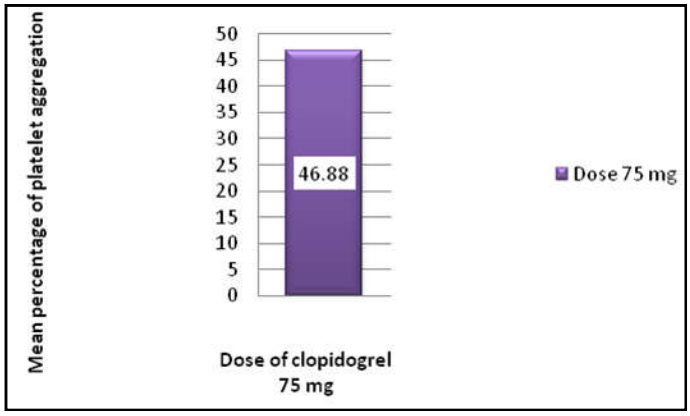

Figure 2. Mean platelet aggregation activity of clopidogrel

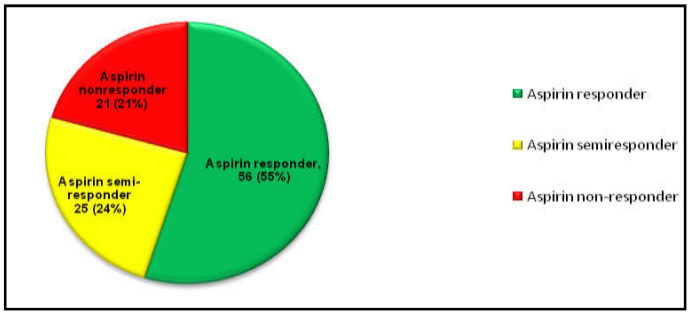

Figure 3. Grouping according to the anti-apatelet aggeregation response response of study subjects on aspirin

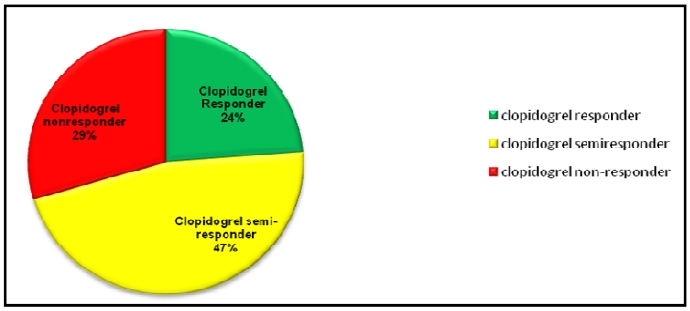

Figure 4. Grouping according to the anti-platelet aggregation response of study subject prescribed clopidogrel

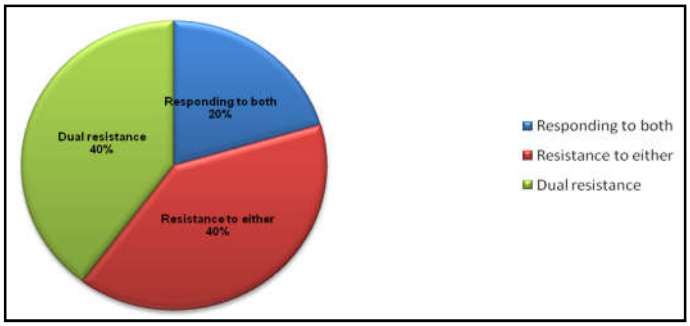

Figure 5. Grouping of the study subjects who took both the drugs according to resistance to the antiplatelet drugs resistance

DISCUSSION

Oral antiplatelet drugs are the mainstay of pharmacotherapy in cardiovascular atherothrombotic diseases. The efficacy of aspirin and clopidogrel via antiplatelet action in decreasing the risk of adverse events in cardiovascular diseases is established since past 20 years. But despite chronic oral antiplatelet therapy, atherothrombotic events continue to occur in number of patients (Feher, 2010). Evidence suggests that a large proportion of patients at high risk of cardiovascular events do not benefit from aspirin monotherapy or aspirin and clopidogrel dual therapy due to numerous factors. One has witnessed the emergence of a new phenomenon of 'aspirin and

clopidogrel resistance'. Thus, a search for platelet function tests that could reliably monitor suppression of antiplatelet targets was initiated (Gasparyan, 2010). Literature reports possible relationships between residual platelet activity and clinical outcomes. This strengthens the possibility of resistance to oral antiplatelet drugs as the underlying cause behind these adverse events (Feher, 2010). Many studies have used different platelet function tests and provided an estimate of prevalence of aspirin resistance ranging from 5.5% to 60% (Gasparyan, 2008) and clopidogrel resistance as 16.8% to 21% (Wang , 2009 and Snoep, 2007). This range is variable in different studies and this prevalence suggests that patients who are taking aspirin and clopidogrel as preventive therapy are still at risk despite being prescribed drugs to protect them. Sharma et al (Sharma, 2009), raised the question of why monitoring wass not being done for platelet function in patients on antiplatelet drugs. They also presented several tests for tailoring antiplatelet therapy and stratified patients into non-responsive, hypo-responsive and responsive to aspirin and clopidogrel. Because of various drawbacks of the platelet function tests, none of the currently available tests have been recommended for clinical practice. Reasons include non-availability of instruments, expensive tests, laborious technique, complex sample preparation, need for large volume of the sample, non-availability of trained staff and many more. Our study tried to evaluate the residual platelet aggregation activity by a "point of care" test based on plateletworks kit (an FDA approved assay) (23) which can be used at places where fewer facilities are available. Our test included taking only 4 ml of blood sample and adding agonist to it at the laboratory and running the sample in impedance based coulter machine (discussed in Material & methods). Though the western literature is flooded with studies on antiplatelet resistance, we could not find any study in India using our methodology. As the definition of resistance to aspirin and clopidogrel is still not well defined we have included non-responders and semi responders in the resistant group of the drugs. A Meta-analysis done showed that the prevalence of laboratory aspirin resistance ranged from 5% to 65%. It had included 12 studies worldwide and pooled them. 1813 patients were included in it. The mean prevalence of aspirin resistance by various laboratory tests was found to be 27% (Snoep, 2007). Sadiq et al (8) evaluated prospectively aspirin resistance in Indian patients with stable coronary artery disease on 150 mg aspirin by platelet aggregometer. In their study aspirin resistance was seen in 2.08% of patients and 39.58% were semi responders. H Mardikar et al (24) studied patients with CAD or stroke or transient ischemic attack or peripheral artery disease or with multiple atherothrombotic risk factors who were receiving aspirin 150 mg daily. In this study 3.1% were said to be hypo-responders.

The prevalence of residual platelet reactivity despite aspirin intake found in our study (45.09%) is close to as shown by Sadiq et al (8) (semiresponder and aspirin resistance-2.08%+39.58%=41.66%). The minor difference may be because the dose of aspirin used in our study was variable (75/150 mg) and doubling of dose of aspirin brings the semi responders to responder group. Also, our study included patients who were follow up cases and hence non-compliance may be a big factor causing variation in the prevalence of resistance. Guha et al (11) assessed both aspirin and clopidogrel resistance in patients with ACS in Indian population and found 17% of patients as aspirin resistant. This included patients within 7 days of initiation of therapy. It is known that patients are more likely to be compliant in this phase of disease as they are hospitalized and medications are being given supervised. Additionally, these cases have recently had a life threatening disease and are more likely to adhere to treatment (25-26). The dose used in their study was 150 mg aspirin. The study subjects in the above studies belonged to urban population while ours was a rural area where compliance differs because lower socioeconomic background, illiteracy etc. It is estimated that 40% of patients with cardiovascular disease do not comply with aspirin (27-29) and poor compliance is said to be an important reason for aspirin being ineffective in the laboratory and clinically settings. Also, aspirin is the 'first-choice-to-stop' drug from an often long list of prescribed treatments (antihypertensives, lipid lowering drugs, antianginals, etc.) in patients with cardiovascular disease (30). Post-MI patients with low educational status are also more likely to discontinue use of all medications (31-32). The prevalence of clopidogrel variable response i.e. clopidogrel resistance in our study was 76.13% which is very high as compared to other studies. A meta-analysis done focusing on clopidogrel resistance with 25 studies and 3688 patients found clopidogrel non-responsiveness at 21%. It also showed that resistance was inversely correlated with time between clopidogrel loading and determination of non-responsiveness and loading dose (14). Kumar et al (9) prospectively evaluated the prevalence of clopidogrel resistance in patients of ACS on dual platelet therapy by platelet aggregometer. Their patients were on clopidogrel 300 mg bolus followed by 75 mg per day for 3 days along with 325 mg aspirin per day. They found that 15.2% showed inadequate response with 2.54% clopidogrel resistant and 12.7% semi-responders. Guha et al (11) found that the clopidogrel resistance was 19% in their study. The prevalence of residual platelet reactivity despite taking clopidogrel was very high in our study as compared to the above studies. The huge difference seen might be because our study included follow up patients and was conducted in rural setting. Clopidogrel takes a long time (2 days) to show its antiplatelet effect even after loading dose so if a patient is skipping medication the effect with clopidogrel will be a lot more variable in comparison with aspirin whose effect is seen within minutes of its intake. The resistant group in our study included semi as well as non-responder in contrast to other studies which have defined their groups differently. Dual antiplatelet drug resistance in our study was found to be 35%. Guha et al (11) found dual antiplatelet drug resistance in India to be 12%. BT Ivandic et al (33) found that 10.4% as dual resistant. In our study, the dual resistance seen was very high as compared to above studies. Reasons might be that most of the patients who are prescribed aspirin and clopidogrel are given so as a fixed drug combination and hence if non-compliance is a factor for this non-reactivity/ resistance, both drugs will not be showing effect. Also, a strong trend toward dual non-responsiveness is seen in patients with ACS. Strong platelet reactivity is frequently found in patients who have acute or severe conditions and cause dual non-responsiveness if alternative pathways of platelet aggregation are up regulated and dominating (e.g. platelet stimulation by thrombin) (Tantry, 2005).

Association of aspirin response with demographic characteristics

Gum et al (Gum, 2003) reported a trend towards increased age in patients with aspirin resistance or semi-responders. Our study didn't show such a trend as the study population was with a mean age approximately 60 years. In our study neither males nor females showed higher aggregation. This was not in coordination with Sadiq et al (2005) and Gum et al (2003) who have reported a higher degree of aspirin non responsiveness in females and Ashwin et al (2007) who found higher aggregation in males. The BMI of patients in both the groups was slightly on higher side with 27.09 ± 3.61 & 26.90 ± 3.76 in both the responder and semi/non-responder group, respectively. In our study, the patients receiving 75 mg of aspirin were less likely to be responder and who received 150 mg of aspirin were more likely to belong to the responder group. The difference in the two groups was statistically significant. Thomson et al (10) showed that overweight patients (who had BMI >24.99) had more aspirin resistance and commented that 75 mg aspirin per day may not be optimal in overweight Indian patient for secondary prevention. Guha et al (2009) showed that they observed a satisfactory inhibition of platelet aggregation after doubling the maintenance dose of aspirin from 150 mg to 300 mg. Thus, suggesting inadequacy of the dose. It suggests that 75 mg of aspirin may not be adequate and patients who are semi-responder while receiving 75 mg of dose might respond adequately if the dose is doubled. Our study supports this fact as in 16 patients who were receiving 150 mg of aspirin per day, only 2 patients showed semi-non responsiveness and 14 responded well to the drug. This also highlights the need for calculating the dose as per body weight or BMI of the patient. If done so, chances of getting full response to the drug increases. But problem in calculation dose and prescribing so is challenged by the availability of low dose aspirin in two doses only, 75 mg and 150 mg. The 100 mg tablet of low dose aspirin should also be available.

Association of clopidogrel response with demographic characteristics: Kumar et al (2002) found that females were more likely to be non-responder but did not find difference with age or BMI. It is also mentioned in literature that in old age there occurs decrease in percentage platelet inhibition with explanation that there occurs age related decrease in drug absorption or in activity of cytochrome P450 3A4, which is essential in the conversion of clopidogrel to its active form. Also old age is associated with more of drug interactions. Guha et al (2009) could not find any association. Our study also like Guha et al (2009) couldn't find any association of age, gender or BMI and sensitivity to clopidogrel. The inclusion criteria was patients on prescription of aspirin and/clopidogrel. So, we could not assure compliance of the patients. Hence, we think non-compliance as a major factor in the variability of responses further adding to the prevalence of resistance to the two drugs. For this further studies should be done to see the platelet function after assuring compliance of the two drugs. Also, physicians should focus on this problem of non-compliance and assure that their patients take their medications as needed. The method of platelet function assay which was an adaptation of plateletworks kit had an advantage that we could collect the sample in sodium citrated bulb and then add the agonists in the lab. In contrast to the kit that it had to be run in coulter machine within 5 min of addition of sample to the test tube as it contained the agonist in the tube itself. So, we got the benefit of time and convenience in our methodology.

Conclusions: Though we got significant results, we still require more insight on this topic to reach a final conclusion. Our study shows that we are definitely facing an upsurge of aspirin and clopidogrel resistance but this problem in India has not gained prominence due to small amount of studies. More studies are required in India as well as in abroad to see the exact prevalence of this problem. The definition of resistance of both the drugs also needs to be universalized. Standard yet less expensive, affordable, specific and sensitive platelet function assays are needed to see the antiplatelet aggregation activity to assure the drugs activity to the fullest. And finally, we should also direct more research in developing more effective antiplatelet drugs so that the problem of resistance and variable responses of aspirin and clopidogrel is overcome and society as a whole is benefited.

REFERENCES

Antiplatelet Trialists' Collaboration. Collaborative overview of randomized trials of antiplatelet therapy. Prevention of death, myocardial infarction, and stroke by prolonged antiplatelet therapy in various categories of patients. *BMJ* 1994; 308(6921):81-106.

Antithrombotic Triatlists' Collaboration. Collaborative meta-analysis of randomized trials of antiplatelet therapy for prevention for prevention of death, myocardial infarction, and stroke in high-risk patients. BMJ 2002; 324(7329):71-86.

Ashwin KA, Bairy KL, Vidyasagar S, Verma M, Prashanth CK, Sachidananda A. Aspirin resistance. Indian J Physiol Pharmacol 2007; 51 (2):109–117.

Carney RM, Freedland KE, Eisen SA, Rich MW, Skala JA, Jaffe AS: Adherence to a prophylactic medication regimen in patients with symptomatic versus asymptomatic ischemic heart disease. Behav Med 1998; 24:35-39.

Cotter G, Shemesh E, Zehavi M, Dinur I, Rudnick A, Milo O, et al. Lack of aspirin effect: aspirin resistance or resistance to taking aspirin? Am Heart J 2004; 147:293-300.

Feher G, Feher A, Pusch G, Koltai K, Tibold A, Gasztonyi B, et al. Clinical importance of aspirin and clopidogrel resistance. World J Cardiol. 2010; 2:171–186.

Gasparyan AY, Watson T, Lip GY. The role of aspirin in cardiovascular prevention: implications of aspirin resistance. *J Am Coll Cardiol.* 2008 May 13; 51(19):1829-43.

Gasparyan AY. Aspirin and clopidogrel resistance: methodological challenges and opportunities. Vasc Health Risk Manag. 2010; 6:109-112.

Guha S, Mookerjee S, Guha P, Sardar p, Deb S, Roy PD, Karmakar R, Mani S, Hema MB, Pyne S, Chakraborti P, Deb PK, Lahari P, Chaudhari U. Antiplatelet drug resistance in patients with recurrent acute coronary syndrome undergoing conservative management. Indian Heart J 2009; 61:348-352.

Guha S, Sardar P, Guha P, Roy S, Mookerjee S, Chakrabarti P, Deb PK, Chaudhari U, Deb S, Karmakar R, Dasgupta AK, Lahiri P. Dual antiplatelet drug resistance in patients with acute coronary syndrome. Indian Heart J 2009; 61:68-73.

Gum PA, Kottke-Marchant K, Welsh PA, et al. A prospective, blinded determination of the natural history of aspirin resistance among stable patients with cardiovascular disease. J Am Coll Cardiol.2003; 41:961–5.

Guyer KE. The present state of aspirin and clopidogrel resistance. Hamostaseologie 2009; 29:285-290.

H. Gerhard Vogel (Ed.). Drug discovery and evaluation: Pharmacological assays, second edition. publisher Springer-Verlag Berlin Heidelberg New York 2008; p. 283-284.

Ho PM, Spertus JA, Masoudi FA, Reid KJ, Peterson ED, Magid DJ, et al. Impact of medication therapy discontinuation on mortality after myocardial infarction. Arch Intern Med 2006; 166:1842-7.

Ho PM, Spertus JA, Masoudi FA, Reid KJ, Peterson ED, Magid DJ, et al. Impact of medication therapy discontinuation on mortality after myocardial infarction. Arch Intern Med. 2006; 166(17):1842-7.

Huffman MD. Coronary heart disease in India. [Full text] 2009 [cited 2011 Nov 06]; Available from: URL:http:// sancd.org/uploads/ pdf/factsheet_CHD.pdf

Ivandic BT, Sausemuth M, Ibrahim H, Giannitsis E, Gawaz M, Katus HA. Dual antiplatelet drug resistance is a risk factor for cardiovascular events after percutaneous coronary intervention. Clin Chem. 2009 Jun; 55(6):1171-6.

Komiya T, Kudo M, Urabe T, Mizuno Y. Compliance with antiplatelet therapy in patients with ischemic cerebrovascular disease. Assessment by platelet aggregation testing. Stroke 1994; 25:2337-42.

Kumar S, Saran RK, Puri A, Gupta N, Sethi R, Surin WR, Dikshit M, Dwivedi SK, Narain VS, Puri VK. Profile and prevalence of clopidogrel resistance in patients of acute coronary syndrome. Indian Heart J. 2007; 59(2):116-7.

Lau WC, Walker CT, Ogilby D, Walsh MM, Carville GM, Guyer BS, et al. Evaluation of bedside platelet function assay: performance and clinical utility. Annals of Cardia Anaesthesia 2002; 5:33-42.

Mardikar H, Deo D, Deshpande N, Mardikar M, Ghosh A, Munot K, et al. Variability in platelet response to a single daily dose of 150 mg enteric coated aspirin in a high risk population. *J Assoc Physicians India.* 2008 May; 56:321-4.

Maree AO, Fitzgerald DJ. Variable platelet response to aspirin and clopidogrel in atherothrombotic disease. Circulation 2007; 115(16):2196-2207.

Matetzky S, Shenkman B, Guetta V, Shechter M, Bienart R, Goldenberg I, Novikov I, Pres H, Savion N, Varon D, Hod H. Clopidogrel resistance is associated with increased risk of recurrent atherothrombotic events in patients with myocardial infarction. Circulation 2004; 109:3171-3175.

Sackett E, Nuttall GA, Ereth MH, Oliver WC. Validation of plateletworks™ point of care platelet function analyzer. Anesthesia and analgesia 2000; S86.

Sadiq PA, Puri A, Dixit M, Ghatak A, Dwivedi SK, Narain VS, Saran RK, Puri VK. Profile and prevalence of aspirin resistance in Indian patients with coronary artery disease. Indian Heart J 2005; 57:658-661.

Schwartz KA, Schwartz DE, Ghosheh K, Reeves MJ, Barber K, Defranco A. Compliance as a critical consideration in patients who appear to be resistant to aspirin after healing of myocardial infarction? Am J Cardiol 2005; 95:973-975.

Shantsila E, Lip GY. 'Aspirin resistance' or treatment non-compliance: which is to blame for cardiovascular complications? J Transl Med. 2008 Aug 29; 6:47.

Sharma RK, Reddy HK, Singh VN, Sharma R, Voelker DJ, Bhatt G. Aspirin and clopidogrel hyporesponsiveness and nonresponsiveness in patients with coronary artery stenting. Vasc Health Risk Manag. 2009; 5:965-72.

Snoep JD, Hovens MM, Eikenboom JC, van der Bom JG, Jukema JW, et al. Clopidogrel nonresponsiveness in patients undergoing percutaneous coronary intervention with stenting: a systematic review and meta-analysis. Am Heart J. 2007 Aug; 154(2):221-31.

Snoep JD, Hovens MM, Eikenboom JC, van der Bom JG, Jukema JW, et al. Clopidogrel nonresponsiveness in patients undergoing percutaneous coronary intervention with stenting: a systematic review and meta-analysis. Am Heart J. 2007 Aug; 154(2):221-31.

Tantry US, Bliden KP, Gurbel PA. Resistance to antiplatelet drugs: current status and future research. Expert Opin Pharmacother 2005; 6:2027-2045.

Tarján J, Salamon A, Jáger R, Poór F, Barczi V, Dinnyés J, et al. The rate of acetylsalicylic acid non-respondents among patients hospitalized for acute coronary disease, previously undergoing secondary salicylic acid prophylaxis. Orv Hetil 1999; 140:2339-43.

Thomson VS, John B, George P, Joseph G, Jose J. Aspirin resistance in Indian patients with coronary artery disease and cardiovascular events. J Postgrad Med 2009; 55:252-6.

Wang TH, Bhatt DL, Topol EJ. Aspirin and clopidogrel resistance: and emerging clinical entity. European Heart Journal 2006; 27:647-654.

Wang ZJ, Zhou YJ, Liu YY, Yu M, Shi DM, Zhao YX, et al. Impact of clopidogrel resistance on thrombotic events after percutaneous coronary intervention with drug-eluting stent. Thromb Res. 2009; 124(1):46-51.

Zhou L, Schmaier AH. Platelet aggregation testing in platelet-rich plasma: description of procedures with the aim to develop standards in the field. Am J Clin Pathol 2005; 123:172–183.

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