Addendum/Corrigendum

1. **The PhD thesis is not certified and signed by guide at the time of the submission.**

Ans: As per the North Bengal University guidelines for submitting thesis, the certificate from supervisor was not attached with thesis but submitted to Registrar's office as a part of record. (Guideline for submitting PhD thesis issued by NBU Registrar: item no.6.

The scholars are requested not to attach copy of the certificate issued by the supervisor and co supervisor if any, in the thesis as the same shall be retained in the office of the Registrar as part of record.)

2. **In Introduction Section, there are a number of references which are cited in text but their details are not mentioned in section 1.7 (page 31) eg. Yello et al. 2007, Jennings 1960, Ishii et al 2005, Holcomb 1963, Pavithron et al 2007 are some of the example.**

Ans: References which are cited in text but their details are not mentioned in section 1.7 (page 31) are given as follows.

There is some typographical error in yello et al 2007, the correct reference is

a) Yellon DM, Hausenloy DJ, Myocardial reperfusion injury, *New Engl J Med. 2007;* 357:1121-1135.

b) Jennings RB, Summers HM, Smyth GA, Flack HA, Linn H. Myocardial necrosis induced by temporary occlusion of a coronary artery in the dog, Arch Pathos. 1960; 70:68-78.

c) Ishii H, Icsimiya S, Kanashiro M, et al. Impact of a single intravenous administration of nicorandil before reperfusion in patients with ST-segment-elevation myocardial infarction, *Circulation.* 2005; 112:1284-1288.

d) P. Pavithran, H Nandeesha, Madan Mohan, Za Charich Bobby, V Sathiapriya, Padnabha Shenoy, Shirdas Sunil and P. Shyma. Dyslipedemia antedates occurrence of clinical hypertension in non- diabetic and non obese male subjects. *Ind. J. Physiology Pharmacol.* 2007; 51: 96-98.

e) Holcomb, G.N. *J.Pharm.Sciences,* 1963; 55: 125.

f) Basheeruddin Asdaq, SM, Prasanna Kumar S. Protective effects of *Semecarpus anacardium* fruit extract against myocardial ischemia reperfusion injury in rats. *Internet J.Alter.Med.* 2009; 7: 1.

g) Zang XF, Tan BK. Anti hyperglycemic and anti oxidant properties of *andrographis paniculata* in normal and diabetic rats.*Clin.Exp.Pharmacol.Physiol.*2000; 27: 358-363.

3. **In page 4, reference of Kloner et al states for 1993 but in reference cited in page 33 reveals it for 1983.**

   Ans: It is a typographical error and should be read as 1983

4. **In the review of Literature, there are some typographical errors.**

   Ans: In page 60 and 61 edaravone is the correct name

5. **The reason for carrying out studies of synthesized derivatives with respect to enzymes and cardiovascular parameters at 10mg/kg (Refer table 5.2 to 5.4) is not clear. Suddenly why antioxidant, anti inflammatory, antipyretic and analgesic studies was carried at 100mg/kg (Table 5.5 to 5.8)? Scientific paper published in Saudi Pharma Journal reveals that three compounds and all activities were studied at 400mg/kg. This anomaly of dose is not clear.**

Ans: A dose of 10 mg/kg b.w. was chosen for cardiovascular studies based on the report of Tsujimoto *et al,* 2005. Though 10 mg/kg b.w. dose is effective in CVS but this dose does not show any significant effect in analgesic, antipyretic activity. Hence 100 mg/kg b.w. was chosen for these activities.

Ref 1: Tsujimoto I, Hikoso S, Yamaguchi O, *et al.* The antioxidant edaravone attenuates pressure overload-induced left ventricular hypertrophy. Hypertension. 2005; 45, 921-6.

Indeed, the analgesic, anti inflammatory and antipyretic activity was not directly related to cardiovascular studies. But some of the research reports like Entman *et al* 1991; Chien GL *et al 1994* have shown the evidence that myocardial injury is associated with inflammatory response and hypothermia. Based on these references the synthesized compounds were screened for analgesic, anti-inflammatory and antipyretic activity.

Ref 2: Entman, ML, Michael LH, Rossen RD, Dreyer WJ, Anderson DC, Taylor A.A, Smith, CW. Inflammation in the course of early myocardial ischemia. *FASEB J.* 1991; 5: 2529–2537.

Ref 3: Chien GL, Wolf RA, Davis RF, Van Winkle DM. "Normothermic range" temperature affects myocardial infarct size.*Cardiovasc Res*1994; 28:1014 -1017.

The peroxides derived from lipid peroxidation such as MDA (TBARS) and 4-HNE have been strongly associated with myocardial ischemic reperfusion injury (Blasig *et al.*, 1995; Ski et al., 2008). Based on this fact, the compounds have been screened for antioxidant activity.

In chronic toxicity study high dose 400 mg/kg was selected since these compounds were safe up to 5000 mg/kg during acute toxicity study. So the dose 400mg/kg was selected for

chronic toxicity study. Moreover, as far as the time is concerned, the chronic toxicity study was limited to three compounds only and the complete study would take several months.

The instrument name to be cited in page 104 where the heart was stored in liquid nitrogen.

Ans: As per the protocol the heart has to be stored in liquid nitrogen but alternatively dry ice was used in my present experiment.

. The % inhibition of rat paw edema (Table 5.6, page 174) needs to be checked as there are large numbers of calculation error in % inhibition of edema in rats.

Ans: The calculation error in % inhibition of edema in rats was detected and submitted herewith after correction. (Table 5.6, page no 174)

. Why was Freund's adjuvant induced polyarthritic model not shown in the thesis though published paper reveals some data?

Ans: Freund's adjuvant induced polyarthritic model was not shown in the thesis because it was done for only three compounds where as Carrageenan induced rat paw edema was done for 10 compounds. Hence it was not included in the thesis.

. The induction of fever by subcutaneous administration of 10% yeast in rabbits is questionable. The rectal temperature should have been measured 4 hrs also after treatment. The reference cited Lu et al (2004) (page 109) is not there in the list.

Ans: As per the procedure given in reference Lu et al 2004, the antipyretic study was done even though the LPS is the suitable pyrogen to induce fever in rabbit.

Lu, W.L., Zhang, Q., Zheng, L., Wang, H., Li, R.Y., Zhang, L.F., 2004. Antipyretic, analgesic and anti inflammatory activities of ketoprofen beta-cyclodextrin inclusion complexes in animals. *Biol. Pharm. Bull.* 27 (10), 1516–1520.

10. In discussion portion (page 187), it is mentioned that all data (paragraph 1 of page 187) are significant at $p<0.05$. But result section (table-5.5, page 171) does not match with the information stated.

Ans: The experimental data were significant at $p<0.05$ for compounds PYZ 8, PYZ 9, PYZ 10 in MDA estimation. But for 4-HNE estimation, the results are significant at $p<0.05$ for compounds PYZ 1, PYZ 5, PYZ 9.

11. The structure activity relationship should be more lucid and a conclusion needs to be made regarding which of ten compounds has maximum activity with least toxicity.

Ans: In addition to SAR mentioned in page 182-183, the following may further be added.

a) The electron donor increases activity as compared to electron withdrawing substituent. This reveals that electron donor (OCH_3) may contribute to increase lipophilicity, which may play very vital role to enhance the biological response.
b) Even though alkoxy (OCH_3), hydroxyl groups are electron donating group, amongst them –OH group substituted pyrazolone is more active than the alkoxy group substituent.
c) The –OH group can facilitate the molecule to undergo keto enol tautomerism without any hindrance.
d) Amongst PYZ 8, 9 & 10 the compound PYZ 10 is more active. It may be due to the presence of hydroxyl group.
e) The increased activity in pyrazolone (PYZ 10) may be due to the capacity to impart keto- enol tautomerism in the molecules.

Based on these SAR, it can be concluded that PYZ 10 is more active amongst the synthesized congeneric series.

: 5.6. Anti inflammatory activity of pyrazolone compounds on carrageenan-induced :dema in rat

›ds /p o	Paw volume in ml, Mean ± SEM (% inhibition of paw edema)			
	1h	2 h	3 h	4 h
rol	0.97 ± 0.016	0.93 ± 0.017	0.91 ± 0.013	0.89 ± 0.013
rin	0.61±0.008(37.11)**	0.53±0.011(43.01)**	0.41±0.008(54.94)**	0.3±0.006(66.29) **
Z1	0.84±0.007(13.40) *	0.65± 0.007 (30.10) *	0.55±0.004 (39.56)*	0.42±0.003(52.80) *
Z2	0.83±0.005(14.43) *	0.67±0.008(27.95)*	0.63±0.005 (30.76)*	0.43±0.003 (51.68) *
Z3	0.88±0.009 (9.27) *	0.68±0.009(26.88)*	0.50 ±0.023(45.05)*	0.411±0.004(53.82)*
Z4	0.88±0.009 (9.27) *	0.68±0.008(26.88)*	0.51 ±0.007(43.95)*	0.40±0.004 (55.05)*
Z5	0.65±0.009(32.98)**	0.55±0.005(40.86)**	0.4 ±0.006(56.04)**	0.30±0.003(66.29)**
Z6	0.69±0.007(28.86)**	0.56±0.006(39.78)**	0.46±0.007(49.45)**	0.31±0.004(65.16)**
Z7	0.65±0.006(32.98)**	0.56±0.008(39.78)**	0.46±0.003(49.45)**	0.38 ± 0.003(57.30)**
Z8	0.73 ± 0.010 (24.74) *	0.61±0.011 (34.40) *	0.51 ± 0.004(43.95)*	0.46±0.005 (48.31) *
Z9	0.75±0.006 (22.68) *	0.66±0.010 (29.03) *	0.53 ± 0.005(41.75)*	0.5 ± 0.006 (43.82)*
Z10	0.63±0.010(35.05)**	0.56±0.015(39.78)**	0.43±0.006(52.74)**	0.36±0.004(59.55)**

:0.01vs Control, * p<0.05vs Control (n=6)

G. Mammyppan

B. P. Sarkar/Supervis