Original Research Article

Biochemical, Histopathological and Mutagenic Changes Following the Co-Administration of Antihelminthic and Antimalarial Drugs in Wistar Rats

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ABSTRACT

Aim

To determine the effects of antimalaria and antheminthic drugs combination in the incidence of histopathological alteration and biochemical modulations in liver and kidney of albino rats.

Place and duration of study

The study was undertaken at the Zoology Department University of Lagos Akoka Lagos Nigeria.

Methodology

A total of twenty (25) Male adult albino rats of 13-15 weeks old were divided into 5 groups of 5 rats each and daily oral administration of human therapeutic doses of praziquantel (PZQ 50mg/kg body weight) separate and in combination with ivermectin (IVM 0.4mg/kg body weight), albendazole (ALB 15mg/kg body weight) and Artemether-lumefanthrine (ACT 140mg/kg body weight) was administered with the group which serve as the control receiving 1ml distilled water. Toxic effects due to these treatments were investigated using histopathological, biochemical and mutagenic indices at day 8th and 15th of the study.

Result

Biochemical assessment revealed significant reduction in AST, ALT, ALP and potassium in the treatment group compared to the control. Increase in the level calcium, Albumin and bicarbonate were also observed in treatment groups. Histopathological assessment of the liver showed a general incidence of focal inflammation along the portal tract area, but did not show any differential severity across treatment groups except for single PZQ treatment group which were characterized by fatty infiltration. A general occurrence of mesangial damage and glomerula injury was observed in kidney tissues. Renal lesions were more severe in single PZQ + IVM treatment groups while mild lesions characterized renal tissue from PZQ+ACT treatment groups. Mutagenic effects as indicated by the high incidence of sperm head abnormalities was recorded across combination treatments especially in PZQ+ IVR and PZQ+ ACT groups.

Conclusion

Findings suggest that combination therapies are synergistic and could result in nephrotoxicity, antidiuretic effects, dehydration and mutagenicity at human therapeutic doses.

Keywords: Nephrotoxicity, Praziquantel, Combination-therapy, Human therapeutic doses, sperm head abnormalities

17 **1.0 INTRODUCTION**

18 The rise in global disease burden has seen an increased therapeutic use of drugs with
19 unknown/poorly understood toxic potential [1]. Many of such implicated drugs include those
20 with adaptable therapeutic applications, which often characterize interventions for public
21 health issues like parasitic infections [2] Recent reports indicate that parasitic and infectious
22 diseases account for about 25% with a bulk of these incidences occurring in Africa,
23 Southeast Asia and Eastern Mediterranean regions [2][3]. Some of the most documented
24 incidences include high incidence of soil-transmitted helminthes infections among children [4]
25 and maternal and infant mortality cases worldwide attributable to malaria annually
26 particularly in Africa [5].
27 Aside fundamental factors like drug availability and costs, current therapeutic use and
28 clinical discretion exercised during the application of antiparasitic drugs are largely guided by
29 the increased incidence of drug-resistant parasites, and the characteristic narrow options of
30 medications for parasitic infections [5][6]. Over time adaptive interventions for helminthic
31 diseases and protozoan infections have included single-dose, safe, and relatively cheap
32 drugs to drugs with a broad-spectrum activity, but with the incidence of drug-resistant
33 pathogen species, elucidation and subsequent insight into the mechanisms underlying
34 intrinsic and acquired drug-resistance has resulted in drug repurposing and development of
35 rational combination therapies to overcome toxicity and resistance [7].
36 The therapeutic administration of drugs and combination therapies have however
37 demonstrated potential for tissue injury or toxicity even when introduced within specified
38 therapeutic ranges [8][1]. Such toxicity may result not only from direct toxicity of the primary
39 compound but also from a reactive metabolite or from an immunologically-mediated
40 response affecting particular cells or tissues [9] which in turn could result in pathological
41 outcomes [10].Other studies have implicated the administration of drug combinations with an
42 increased production of Reactive Oxygen Species (ROS) [11]. Post-drug intake effects in
43 organs have been a key strategy for monitoring and determining drug-related toxicities [12]. It

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44 is against this background that this study investigated the role of antimalaria and
45 anthelminthic drug combinations in the incidence of histopathological alterations and
46 biochemical modulations in liver and kidney of Albino rats and also observing possible
47 mutagenic changes.

48 2.0 MATERIALS AND METHOD

49 2.1 Test animals

50 A total of twenty-five (25) male adult albino rats (*Rattus norvegicus)* Wistar strain of 13-15
51 weeks old with an average weight of 180g±20 were used for the studies. The animals were
52 purchased from an animal farm located in Ikorodu Lagos Nigeria and were maintained in the
53 laboratory for 15 days with cross ventilation at controlled room temperature (27±2°C) and
54 relative humidity (40-60%) with a 12-hour light and dark cycle to acclimatize in the laboratory
55 before the commencement of exposure period. All the rats were housed in conventional
56 plastic cages. These standard cages were bedded with dry wood shavings, which were
57 changed every 2 days to prevent maggotry. The animals were provided daily with fresh
58 supply of standard feeds weighing 150g and water *ad libtum*.

59 2.2 Drug treatment and Sample preparations

60 Praziquantel (PZQ), Albendazole (ALB), Ivermectin (IVM) and Artemether-Lumefantrine
61 (ACT) were used for study. The praziquantel tablet manufactured by BDH industries limited
62 Mumbai india was purchase from a local pharmacy in Lagos Nigeria. Ivermectin Mectizan® a
63 product of Merck& Co., Inc., Whitehouse station, New Jersey, USA was obtained from D-hub
64 pharmacy Ikeja. Albendazole (Zentel) manufactured by SmithKline Beecham laboratories
65 pharmaceuticals France and Artemether-Lumefantrine (Lonart Ds) manufactured by Bliss
66 GVS pharmacy limited India was purchased from the University of Lagos community
67 pharmacy. The drugs were grounded separately with mortar and pestle, weighed and
68 measured at different concentration depending on the mean body weight of the experimental
69 groups.

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70 **2.3 Experimental Design**

71 Before exposure physical parameters such as laboratory temperature and humidity was
72 determined. The human therapeutic dose for each drugs PZQ, IVM, ALB and ACT are
73 50mg/kg, 0.4mg/kg, 15mg/kg, and 140mg/kg body weight respectively. For the experiment
74 there were 5 groups containing 5 rats (Table 1).

75
76 **Table 1: Exposure group and treatments**

S/N	GROUPS	MEAN WEIGHT OF RATS (g)	DRUG ADMINISTERED
1	Control	141.2	1ml distilled water
2	PZQ alone	182.4	Praz 9.12mg
3	PZQ + IVM	190	Praz 9.5mg +Ivr 0.08mg
4	PZQ+ ALB	166	Praz 8.3mg + Abz 2.49mg
5	PZQ + ACT	147.8	Praz7.39mg +ACT 20.7mg

77

78 **2.4 Drug administration**

79 The administration of drugs commenced 15 days after acclimatization as described by Ismail
80 *et al* [13] using oral route for 15 days for all groups except for group 5 in which ACT was
81 administered at the last 3 days of exposure, after which they were sacrificed 24hrs after the
82 last dose was administered based on the methodology by [14]. Animals were weighed after
83 acclimatization on the first day of exposure and the record served as the initial body weight
84 (Day 0). The procedure was repeated on the 8th day of exposure and before sacrificing at the
85 expiration of the required time of exposure and value obtained served as the final body
86 weight. The animals were observed daily for any clinical sign or behavioral changes.

87 **2.5 Collection of blood and tissues**

88 Blood specimen was collected in lithium heparin bottles and fluoride oxalate bottles. Liver,
89 kidney and the cauda epididymis were excised. The cauda epididymis was used for

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mutagenicity examination. The internal organs were placed in a plain bottle and Buoin's fluid added to preserve the specimen for histopathological examination.

2.6 Biochemical analysis

The method according to [15][16][17]was used to determine the biochemical parameters. Blood sample collected during heart excision of rats was used for quantitative determination of protein, albumin, alanine aminotransferase (ALT), aspartate aminotransferase (AST) and alkaline phosphatase (ALP), urea, total bilirubin and creatinine, inorganic phosphate, cholesterol, fasting glucose, Na^+, K^+, Ca^{++} and Cl^- using standard kits.

2.7 Histological Preparations

Representative liver tissue of each group was excised, trimed of fat and other connective tissue and prepared for histological studies. The tissue samples were fixed using 10% formaldehyde solution (formalin) for 24 hours and were later transferred into alcohol to remove excess water. This was followed by the application of a hydrophobic clearing agent, xylene to remove the alcohol before embedding with molten paraffin wax, the infiltrating agent which replaces the xylene. After the tissues have been dehydrated, cleared and infiltrated with the embedding material, they were placed into molds along with liquid materials (wax) which was then hardened. Microtome was then used to cut the paraffin wax embedded tissues. A steel knife mounted in the microtome was used to cut a 10 micrometer thick tissue section which was mounted on a glass microscope slide. The sections samples were stained with haematoxylin and eosin. The slides were examined under CX21 Olympus microscope of magnification of 40X objective and their photomicrograph taken with a Canon (Meville, NY) Power Shot G2 digital camera.

2.8 Mutagenicity Assay

Mutagenicity was determined from sperm head abnormalities. Four (4) male rats were sacrificed for each group by cervical dislocation after anesthetization. The cauda epidydimis

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116 excised from the male rat were placed in a Petri-dish containing 1ml of physiological saline
117 and then minced and teased carefully well with fine scissors and forceps to release the
118 spermatozoa. After gentle pipetting, the suspension is separated from the tissue fragments
119 and a drop of 1% Eosin Y solution in the ratio (10: 1) was added to the suspension for 30
120 minutes. Air-dried smears were prepared on clean, grease-free glass slides using another
121 clean slide angularly positioned at 45° to spread the drop through the whole length of the
122 slide. The slides were then coded, randomized and cytologically examined under a binocular
123 light microscopy with 400x magnification. Sixteen separate slides were prepared for each
124 group for sperm examination. For each group, 2000 sperm cells were assessed for
125 morphological aberration according to the criteria of [18]. The percentage abnormality of the
126 sperm cells in the rats was calculated by using the mean value of the group.

127 $$\% \text{ abnormality} = \frac{\text{Total no of abnormal sperm cells}}{\text{Total no of sperm cells}} \times 100$$
128

129 **2.9 Statistical Analysis**

130 All data were expressed as mean ± standard deviation. One-way analysis of variance
131 followed by Dunnett T_3 post hoc test was used for determining the statistical significance of
132 the data. A probability level of less than 5% ($p<0.05$) was considered significant in all
133 instances. All statistical tests were performed with SPSS 21 version package and originlab
134 version 9.0.

135
136
137 **3.0 RESULTS**
138
139 **3.1 Weight change across drug-treatment groups**

140 Change in weight of experimental animals was assessed at 8th and 15th day intervals during
141 the treatment period. Findings showed that exposure groups showed the highest weight
142 change occurred in the drug-treatment groups particularly in single praziquantel exposure
143 and Albendazole combinations. Both treatment groups showed higher significant weight

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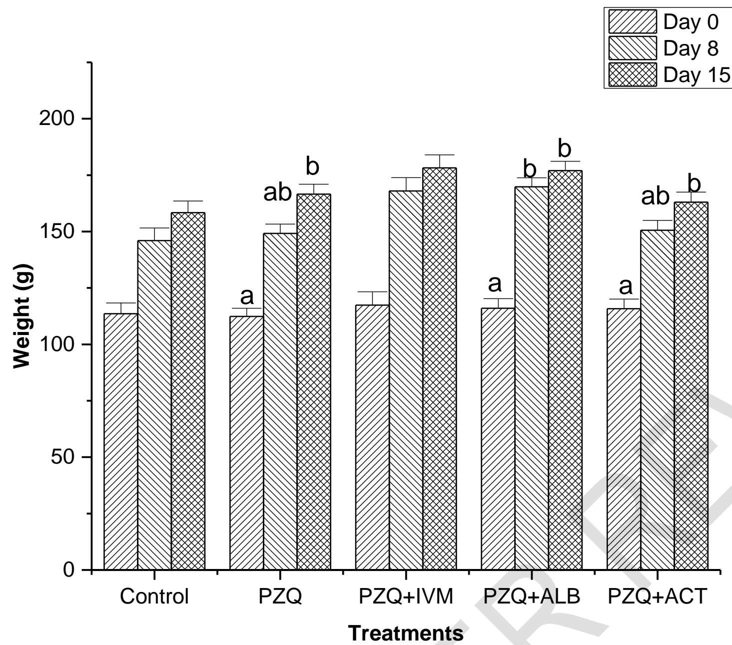
144 difference at the beginning and end of the experiment when compared to control and Praz +
145 ACT treatment group (Figure 1).



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147 **Figure 1: Weight change across control and drug treatment groups (bars within the same group**
148 **with the same alphabet are not significantly different, where error bar=standard error)**
149

150 **3.2 Histopathology**

151
152 Histopathology for liver on slides C, D, E, show focal inflammation with subtle features of
153 hepatocyte loss. Appearance of these cells suggests focal loss which can be through
154 apoptosis/necrosis (Figure 2).

155 For the kidney sample, Plate A which is the control showed subtle features of lobulation of
156 the glomeruli. While Plates B and C i.e. PZQ and PZQ+IVM administered rats respectively
157 showed significantly higher levels of severity compared to plates D and E (Figure 3). Levels
158 of severity compared to plates D and E (Figure 3).

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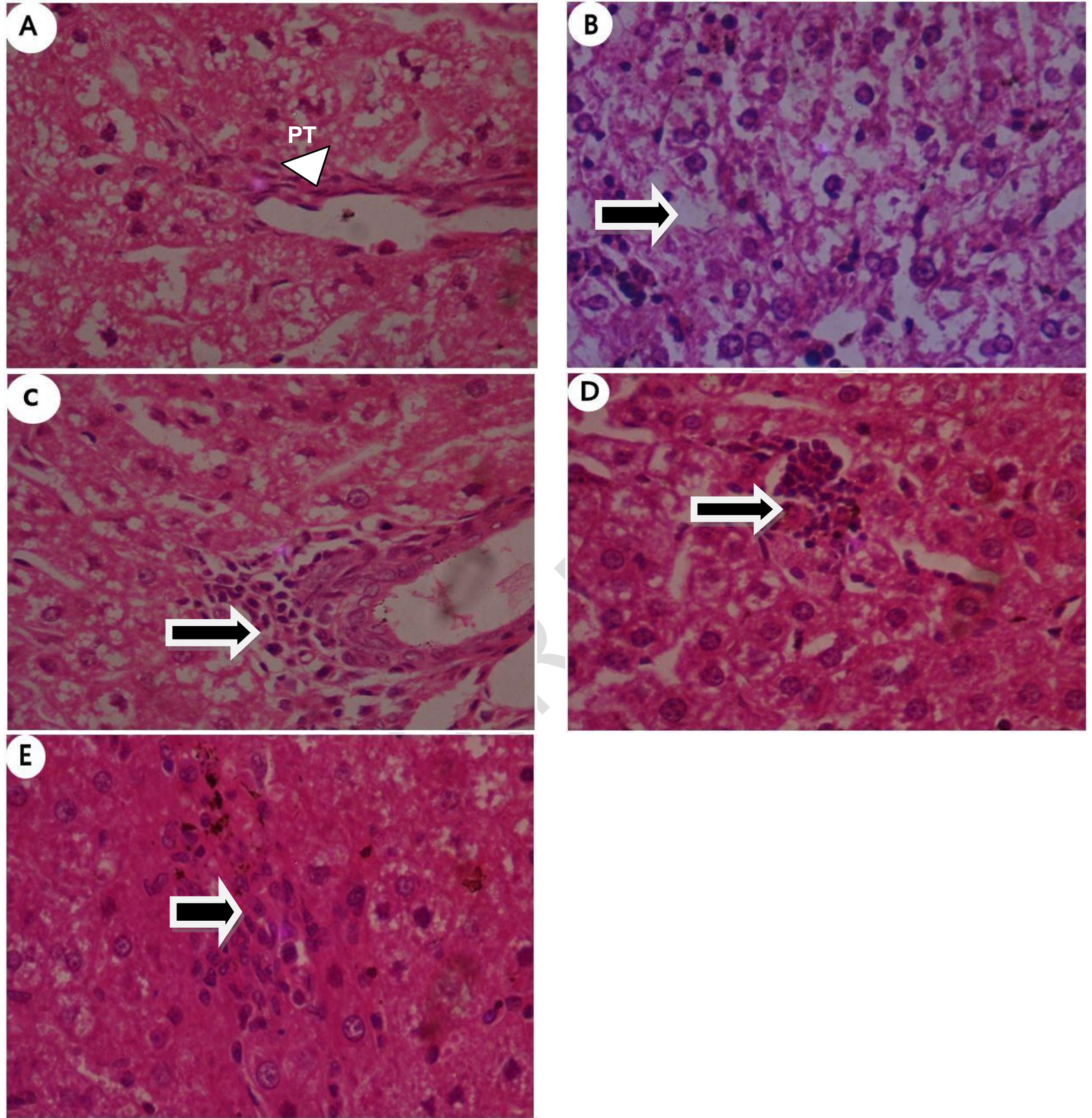

160 **Figure 2: A: Histological section of liver tissue of control albino rats showing normal hepatocytes, bi-**
161 **nucleated cells, cytoplasm and nucleus surrounded by a nuclear membrane and nucleolus around the**
162 **portal tract area (PT) (arrow head) (Magnification X40) B: Histological sections of liver of albino rat**
163 **administered PZQ showing fatty infiltrations (long arrow) C: Histological section of liver of albino rat**
164 **administered PZQ+IVM showing focal inflammation (long arrow) around hepatic portal tract area with subtle**
165 **features of hepatocyte loss. D: Histological sections of liver tissue of albino rat administered PZQ+ALB**
166 **showing focal inflammation (long arrow) with subtle features of hepatocyte loss. E: Histological section of**
167 **liver tissue of albino rat administered PZQ+ACT showing focal inflammation (short arrow) with subtle**
168 **features of hepatocyte loss.**

169

170 **3.3 Biochemical Analysis**

171 Liver enzyme profile across experimental groups showed that at least one treatment group
172 have significantly lower levels of AST, ALT and ALP (Table 2). Also result of analysis
173 showed that drug-treatment groups showed significantly higher levels of albumin compared
174 to the control while creatinine was higher in serum of control animals. Other biomolecule
175 variables such as glucose, urea and cholesterol did not differ significantly between drug-
176 treatment groups and control (Table 2).

177 Electrolyte profile analysis depicted that ALB+IVM combination treatments showed
178 significantly higher levels of sodium ion in serum compared to the control, while all treatment
179 groups showed significantly lower levels of potassium ion compared to the control. All
180 treatment groups showed significant elevated levels of calcium ion in serum compared to the
181 control while all treatment groups except the ACT combination treatment group showed
182 significantly lower levels of phosphate ion in serum compared to the control. Bicarbonate ion
183 levels were significantly elevated in treatment groups compared to the control while
184 significant loss of chlorine ion in serum was recorded in the IVM drug-treatment group
185 compared to the control.

186 **Table 2:Analysis of biochemical variables in rats from control and PZQ, PZQ+IVM,**
187 **PZQ+ALB and PZQ+ACT treatment groups.**

Drugs exposure	AST (µ/L)	ALP (µ/L)	ALT (µ/L)	GLU (mmol/l)	UR (mmol/l)	ALB (mmol/l)	CRE (mg/d)	CHO (mmol/l)
Control (µ/L)	94.40 ± 27.07	44.64±10.59	32.00±14.93	5.32 ±0.83	5.10 ±3.09	37.46±2.8	60.60±7.3	2.10±0.4
PZQ (µ/L)	44.60 ± 15.13	33.14 ± 7.58	22.80 ±7.67	5.96 ±2.38	6.58 ±1.37	40.00±2.9	52.92±3.1	2.18±0.2
PZQ+IVM (µ/L)	48.50 ± 13.17	42.25 ±4.22	23.50 ±12.38	5.40 ±3.16	4.50 ±2.57	32.04±174	39.52±2236	1.82±1.6
PZQ+ALB (µ/L)	44.60 ±13.09	38.84 ±3.81	20.80 ±2.59	5.16 ±0.59	5.89 ±1.07	39.98±0.8	51.82±4.32	2.36±0.7
PZQ+ACT (µ/L)	57.40 ± 9.13	41.42 ±4.88	25.20 ±1.79	5.06 ±1.25	6.36 ±1.16	38.44±2.6	52.18±2.27	2.32±0.9

188 AST=Aspartate aminotransferase, ALP=Alanine phosphatase ALT=Alanine aminotransferase GLU=Glucose,
189 UR=Urea, ALB=Albumin, CRE=Creatinine, CHO=Cholesterol
190
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Table 3: Electrolyte variables in rats from control and PZQ, PZQ+IVM, PZQ+ALB and PZQ+ACT treatment groups.

Concentration of serum electrolytes	Control (µ/L)	PZQ (µ/L)	PZQ+IVM (µ/L)	PZQ+ALB (µ/L)	PZQ+ACT (µ/L)	Reference values
Na^+(mmol/l)	142.9 ±5.52	142.6±1.22	115.6±6.66	143.7 ±1.01	142.0 ± 2.35	144.33meq/l
K^+(mmol/l)	7.39 ±1.95	5.32 ± 0.62	4.73 ±2.67	5.75 ± 0.36	5.83 ± 0.58	5.26meq/l
Ca^{2+} (mmol/l)	1.68 ± 0.09	1.99 ± 0.22	1.69 ± 0.98	1.87 ± 0.18	1.88 ± 0.13	10.17mg/dl
PO_4^{2+}	1.42 ±0.13	1.07± 0.25	1.04 ±0.05	1.31± 0.34	1.24± 0.22	1.8-2.3
HCO_3(mmol/)	12.00 ±4.79	15.40±3.91	12.00 ±7.04	14.20 ± 2.59	14.00 ±4.64	18-30
Cl^{2+}	102.0 ± 2.83	100.4±1.82	82.00 ± 4.8	102.6 ± 2.70	103.6 ±1.52	103.75meq/l

Na^+= Sodium, K^+= Potassium,Ca^{2+}= Calcium, PO_4^{2+}= Phosphate, HCO_3= Bicarbonate
Cl^{2+}= Chloride

3.4 Sperm head abnormality assessment

Five different forms of sperm head abnormality were observed in the rat during the *in vivo* evaluation of the drugs. These include pin head (most prominent), no hook, hook at wrong angle, amorphous and bent sperm. The pin head sperm abnormality appeared predominantly in both the control and exposed group.

Table 4; Showing abnormal sperm cell recorded in experimental rats across treatment and control groups

GROUP	I.D	Normal	Amorphous	Bent	Wrong angle	No Hook	Pin head	Mean abnormal sperm cell	Mean no. of sperm cells	% Abnormal sperm
CONTROL	1	1670	30	45	-	-	-	68.25±49.14[a]	1751.8 ± 52.43[a]	3.8
	2	1708	30	7	28	-	38			
	3	1721	2	-	-	-	2			
	4	1635	-	-	-	-	60			
PZQ	1	1773	64	-	-	-		356.5±246.2[a]	1677.0±226.38[a]	21.25
	2	1384	44	-	40	-	250			
	3	680	176	395	30	-	65			
	4	1445	67	-	135	-	160			
PZQ+IVM	1	1281	209	123	123	-	238	710.8±182.3[b]	1561.8±607.32[a]	45.51
	2	689	210	175	-	-	270			
	3	235	35	154	20	-	20			
	4	1199	82	144	292	25	258			

PZQ+ALB	1	640	472	-	14	-	-	581.3±126.8[ab]	1397.8±254.52[a]	41.58
	2	1026	281	4	276	-	151			
	3	869	233	15	118	-	94			
	4	731	230	89	43	-	305			
PZQ+ACT	1	731	180	134	37	-	6	809.0±410.9[ab]	1676 ± 498.01[a]	48.26
	2	972	267	224	175	-	342			
	3	849	45	240	-	-	313			
	4	908	551	247	91	-	468			

208

209

210 **3.5 DISCUSSION**

211 A number of drugs with poorly understood scope of toxicity currently constitute drug options
212 for public health interventions, particularly for parasitic diseases which have a high incidence
213 among developing nations [5][19].As such a necessary step to avert drug-related biochemical
214 disruption, pathological outcomes and mutagenic effects is to adequately examine and
215 profile the toxic potential of drugs commonly used for public health interventions [1][20].

216 The biochemical modulations observed across drug-treatment groups represented in this
217 study presents very interesting findings. The characteristic concurrent increase in albumin
218 and calcium in both single PZQ drug treatment and PZQ+IVM treatment groups strongly
219 highlight dehydration of animals in the both group. Dehydration has been implicated as a
220 common cause of mild or transient hypercalcemia because when there is less fluid in the
221 blood calcium concentrations rise [21]. The possibility of dehydration was also confirmed from
222 the PCA where a negative correlation between PZQ, PZQ+IVM groups and serum
223 phosphate was depicted. The negative correlation suggests hypophosphatemia which could
224 also be diagnostic for dehydration. Also from the PCA, the positive relationship between
225 albumin and Ca in these treatment groups could be explained on the basis that albumin
226 binds calcium, thyroid hormones, fatty acids, and many drugs, keeping them in the blood
227 circulation and preventing them from being filtered out by the kidneys [22]. The importance of
228 albuminin the effectiveness and toxicity of therapeutic drugs and in drug interactions has
229 been documented [22]. Furthermore, the negative correlation of these treatment groups with
230 chloride (Cl^-) indicates decreased chloride levels in serum of these drug treatment groups.

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231 This decrease in serum chloride levels is diagnostic of tendencies towards hypochloremic
232 alkalosis. Since this is an acute drug treatment study, could be described as acute
233 hypochloremic alkalosis.

234 On the other hand, the strong negative correlation between the PZQ+ALB drug treatment
235 group with potassium ion is suggestive of hypokalemic tendencies, while its positive
236 association with sodium ion highlights hyponatremia which is also suggestive of dehydration.
237 The combination of these two conditions highlights possibilities of metabolic alkalosis.
238 Studies have shown that the kidneys compensate for loss of potassium by retaining sodium
239 in the collecting ducts at the expense of hydrogen ions (sparing sodium/potassium pumps to
240 prevent further loss of potassium), leading to metabolic alkalosis [23][24]. The strong positive
241 correlation between the PZQ+ALB treatment group and bicarbonate levels confirms the
242 possibilities of metabolic alkalosis [25]. Although this altered electrolyte levels may can be
243 attributed to the drug treatments, such patterns of electrolyte alterations may imply severe
244 deleterious outcomes to patients with individual physiological risk factors e.g. advanced age,
245 hypertension, gout and hyperuricaemia, diabetes mellitus, chronic renal failure and use of
246 diuretics. Hypercalcaemia observed in single PZQ and combinations with IVM has been
247 reported to enhance nephrotoxic drug injury by inducing pre-renal physiology [11]. Metabolic
248 alkalosis which was also diagnosed in the treatment groups can result in alkaline urine which
249 increases precipitations of drug crystals within the tubular lumen of the kidney [11][26]. In
250 general, it was inferred that the single and combination PZQ treatment groups except
251 PZQ+ACT demonstrated anti-diuretic symptoms and tendencies towards metabolic
252 disruptions.

253 Although focal necrosis and inflammation of portal tract were common features across all
254 PZQ drug combination, the absence of gradient or severity across treatment groups highlight
255 one of the non-specific possibilities of histopathological assessment. Studies have noted that
256 drug-related injury can mimic all the patterns observed in primary liver disease, making

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257 unequivocal histological diagnosis difficult or almost impossible in the majority of the case [9].
258 Findings from this study juxtaposed with relevant literature indicates that the PZQ
259 combination treatment groups were likely to depict incidence of acute hepatitis.
260 Ramachandra and Kakar [9] noted in their review of drug-induced liver disease that one of
261 the hallmarks of acute hepatocellular injury are portal and parenchymal inflammation,
262 hepatocellular injury and/or necrosis. Foci of inflammatory cells have been reported to occur
263 spontaneously in livers of rodents in prechronic studies [27]. Other studies have also
264 confirmed that inflammatory cell aggregates may be accompanied by evidence of
265 hepatocellular necrosis [28][29].

266 The fatty infiltration (steatohepatitis or steatonecrosis) observed in liver tissues from the PZQ
267 treatment demonstrates onset of liver degeneration. Drugs or their metabolites could inhibit
268 esterification of fatty-acid within the hepatocyte resulting in hepatic vesicles engorged with
269 fatty acids [30]. Such drug-related incidences have been reported for alcohol i.e. alcoholic
270 fatty disease [31] tetracycline [32] and Sodium valproate [33].

271 The more distinct pathology observed in kidney tissues across drug-treatment groups. This
272 trend is expected because pharmacokinetic studies of PZQ reveal that in spite of the large
273 absorption that occurs within the gastrointestinal tract (about 80%), only a relatively small
274 amount enters systemic circulation due to extensive first-pass metabolisms. As a result, PZQ
275 and its metabolites are mainly excreted renally within 24 h after a single oral dose, 70 to
276 80% is reportedly found in urine, but less than 0.1% as the unchanged drug [34][35]. This
277 implies that PZQ will have more interaction with the kidney compared to the liver. Reports
278 have shown that the role of the kidney as a primary eliminator of exogenous drugs and
279 toxins makes it vulnerable to develop various forms of injury [20].

280 Furthermore, the realization that PZQ is metabolized through the cytochromeP450 pathway
281 via CYP3A4 also highlights risks for the kidney. This is because CYP450 which constitutes
282 part of the renal enzyme systems favours the formation of toxic metabolites and reactive

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283 oxygen species [36][35][37]. The presence of these by-products of metabolism tilts the balance
284 in favour of oxidative stress, which outstrips natural antioxidants and increases renal injury
285 via nucleic acid alkylation or oxidation, protein damage, lipid peroxidation and DNA strand
286 breaks [36][38].

287 The mild mesangial damage in single PZQ drug treatment group compared to the severe
288 mesangial damage in PZQ+IVM and PZQ+ALB treatment groups, suggests that ivermectin
289 and albendazole could enhance renal toxicity. Incidence of proximal cell tubular toxicity is
290 indicative of drug-induced nephrotoxic effects e.g. phospholipid damage, increased
291 intracellular calcium concentrations. Other effects include osmolar effects with loss of normal
292 cell contact and tubular occlusion [37][39].

293 The mutagenicity test as indicated by the occurrence of sperm head abnormality, recorded
294 high incidence of abnormality in all drug treatment groups. The higher incidence of
295 abnormality in PZQ+ IVM and PZQ+ACT were statistically significant ($P<0.05$). The
296 predominance of pinhead sperms over all other varying types of sperm head abnormality in
297 the treated groups is consistent with reports on PZQ administered to albino mice for a period
298 of 5-8 weeks [40]. The non-significant difference in incidence of sperm head abnormalities
299 between the control group and PZQ treatment group confirms early reports on the non-
300 mutagenic potential of PZQ treatments in humans [41][42]. Considering the non-mutagenic
301 effects of PZQ demonstrated from this study and the non-mutagenic potential of IVM earlier
302 reported [43], mutagenic effects of combination therapies may be attributed to synergistic
303 interaction of the drugs.

304

305 **4.0 CONCLUSION**

306 Identifying drug-related risks and drug-induced injury is the key to reducing risk of damage to
307 vital organs as liver and kidney. Findings from this study depict that single praziquantel
308 administration and combinations with Ivermectin and albendazole at human therapeutic
309 doses portends risks of liver inflammation, while combination treatments are most likely to

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induce metabolic disruptions, antidiuretic effects and likelihoods of weight gain due to dehydration. Combination treatments are also likely to induce mutagenic effects as indicated by higher incidence of sperm head abnormalities.

Since drug-related risk factors are one of many factors that influence liver and kidney toxicity, more extensive profiling of common drugs options for public health interventions is recommended. This will inform clinical decisions that could increase the risk factors and deleterious outcomes of patients.

COMPETING INTERESTS

There is no competing interest among authors.

CONSENT

Not applicable.

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