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Research Article

Blood Na^+/K^+ and Cl^- Levels of Hyperglycemic Rats Administered with Traditional Herbal Formulations

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ABSTRACT

Background/Aim: The present study investigated alterations in blood Na^+/K^+ and Cl^- levels of alloxan-induced hyperglycemic rats (HyGR) administered with single and combinatorial herbal formulations of leaf extracts of *Acanthus montanus*, *Emilia coccinea*, *Hibiscus rosa sinensis* and *Asystasia gangetica*. **Materials and Methods:** Serum electrolytes concentrations were measured using the ion selective electrode analyzer. **Results:** Serum Na^+ concentration (SNC) of the untreated HyGR (DIAB group) was significantly ($p<0.05$) lower than that of the normal control rats (NORM group).However, HyGR administered with single herbal formulations (SHf) exhibited relatively higher SNC than that of the DIAB group ($p< 0.05$), which was within the range of 133.3 ± 13.6 mMol/L–138.2 ± 15.3 mMol/L; $DIAB_{[Na+]}=127.8\pm12.4$ mMol/L. Serum K^+ concentration (SKC) of the NORM group was significantly ($p<0.05$) higher than that of the DIAB group. Serum Cl^- concentration (SCC) of the DIAB group was not significantly different ($p>0.05$) from those treated with SHf. Also, SCC of HyGR treated with triple herbal formulations (THf) was not significantly different ($p>0.05$) from HyGR treated with double herbal formulations (DHf), THf and quadruple herbal formulation (QHf). SNC exhibited a strong positive correlation with SCC ($r=0.703231543$). Conversely, SNC versus SKC ($r=0.386096385$) and SKC versus SCC ($r=0.454718488$) gave weak positive correlations. **Conclusion:** The present study showed that the various herbal formulations, in the present form, exhibited varied limited capabilities but substantial potentials to reversing altered blood electrolytes levels of HyGR within the 30-day treatment. Furthermore, definitive blood Na^+/K^+ and Cl^- levels could be of diagnostic relevance in the course of management and treatment of DM.

Key words: Chloride, Electrolytes, Herbal formulation, Potassium, Sodium.

INTRODUCTION

Sodium (Na^+), for the most part, is associated with chloride (Cl^-) and bicarbonates (HCO_3^-) in the regulation of acid-base balance of body fluids.[1, 2] Also; extracellular Na^+ plays a major role in the maintenance of osmotic equilibrium as well as uniform distribution and conservation of body fluids. Potassium (K^+) is the principal intracellular cation responsible for neuromuscular excitability, maintenance of acid-base balance, cardiac action and acts as cofactor for pyruvate kinase activity.[1] Studies have shown that the renal system conserves Na^+ more efficiently than K^+.[2]

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Alterations in electrolytes homeostasis may be the outcome of physiologic disorders associated with compromised renal function,[3,4] vomiting and dehydration.[5,6] Additionally, Na^+/K^+-ATPase activity is regulated by insulin action.[7,8] Therefore, low circulating insulin level compromises Na^+/K^+-ATPase activity with concomitant poor Na^+ and K^+ metabolism and transport across biomembranes as well as hindered facilitative uptake of monosaccharides by intestinal epithelia.[9,10] In diabetes mellitus (DM), hyperglycemia imposes glucose induced osmotic diuresis with resultant loss of body fluids and

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electrolytes. The failure of DM patients to conserve body fluids and electrolytes is often exacerbated by underlying renal dysfunction and certain orthodox medications used for the management of DM[11,12] which elicit alterations in electrolytes homeostasis.[6,13,14]

In African and Asian traditional medicinal practices, extracts from *Acanthus montanus*, *Emilia coccinea*, *Hibiscus rosa sinensis* and *Asystasia gangetica* are used for the alleviation of several pathologic conditions.[15-20] Furthermore, the profiles of bioactive principles of the four medicinal plants have been previously reported elsewhere,[17,19-22] in which the presence of phytochemicals such as the alkaloids, tannins, glycosides, carbohydrates, flavonoids and steroids in these plants were responsible for their therapeutic actions. The present study investigated alterations in blood Na^+/K^+ and Cl^- levels of alloxan-induced hyperglycemic rats (HyGR) administered with single and combinatorial herbal formulations of leaf extracts of *A. montanus*, *E. coccinea*, *H. rosa sinensis* and *A. gangetica*.

MATERIALS AND METHODS

Collection and preparation of herbal samples

Fresh leaves of *Acanthus montanus* (Nees) T. Anderson (ACMO), *Emilia coccinea* G. Don (EMCO), *Hibiscus rosa sinensis* L. (HIRO) and *Asystasia gangetica* L. T. Anderson (ASGA) were collected between the months of July and August, 2009. Preparation of the herbal extracts was according to the methods previously described.[23]

Induction of hyperglycemia/experimental design

Induction of hyperglycemia was as previously described.[24] Animal handling was in conformity with the standard principles of laboratory animal care of the United States National Institutes of Health (NIH, 1978). A total of 102 male Wistar rats were allotted into seventeen (17) groups of six (6) rats each. The animal groups were designated on the basis of treatments received at regular intervals of 2 days for 30 days. Herbal treatments of the HyGR were described as single herbal formulations (SHf), double herbal formulations (DHf), triple herbal formulations (THf), and quadruple herbal formulation (QHf).

- NORM: Normal rats received standard commercial feed (SCF) + water *ad libitum* + 1.0 mL/kg of PBS.
- DIAB: HyGR received SCF + water *ad libitum* + 1.0 mL/kg of PBS.
- SHf-ACMO: HyGR received SCF + water *ad libitum* + *A. montanus* (20 mg/kg in PBS; i.p.).
- SHf-ASGA: HyGR received SCF + water *ad libitum* + *A. gangetica* (20 mg/kg in PBS; i.p.).
- HrEMCO: HyGR received SCF + water *ad libitum* + *E. coccinea* (20 mg/kg in PBS; i.p.).
- HrHIRO: HyGR received SCF + water *ad libitum* + *H. rosa sinensis* (20 mg/kg in PBS; i.p.).
- DHf-AGAM: HyGR received SCF + water *ad libitum* + combined dose (ratio: 1:1 *w/w*) of *A. gangetica* + *A. montanus* (20 mg/kg in PBS; i.p.).
- DHf-AGEC: HyGR received SCF + water *ad libitum* + combined dose (ratio: 1:1 *w/w*) of *A. gangetica* + *E. coccinea* (20 mg/kg in PBS; i.p.).
- DHf-AGHR: HyGR received SCF + water *ad libitum* + combined dose (ratio: 1:1 *w/w*) of *A. gangetica* + *H. rosa sinensis* (20 mg/kg in PBS; i.p.).
- DHf-AMEC: HyGR received SCF + water *ad libitum* + combined dose (ratio: 1:1 *w/w*) of *A. montanus* + *E. coccinea* (20 mg/kg in PBS; i.p.).
- DHf-AMHR: HyGR received SCF + water *ad libitum* + combined dose (ratio: 1:1 *w/w*) of *A. montanus* + *H. rosa sinensis* (20 mg/kg in PBS; i.p.).
- DHf-ECHR: HyGR received SCF + water *ad libitum* + combined dose (ratio: 1:1 *w/w*) of *E. coccinea* + *H. rosa sinensis* (20 mg/kg in PBS; i.p.).
- THf-AGEH: HyGR received SCF + water *ad libitum* + combined dose (ratio: 1:1:1 *w/w*) of *A. gangetica* + *E. coccinea* + *H. rosa sinensis* (20 mg/kg in PBS; i.p.).
- THf-AMAE: HyGR received SCF + water *ad libitum* + combined dose (ratio: 1:1:1 *w/w*) of *A. montanus* + *A. gangetica* + *E. coccinea* (20 mg/kg in PBS; i.p.).
- THf-AMAH: HyGR received SCF + water *ad libitum* + combined dose (ratio: 1:1:1 *w/w*) of *A. montanus* + *A. gangetica* + *H. rosa sinensis* (20 mg/kg in PBS; i.p.).
- THf-AMEH: HyGR received SCF + water *ad libitum* + combined dose (ratio: 1:1:1 *w/w*) of *A. montanus* + *E. coccinea* + *H. rosa sinensis* (20 mg/kg in PBS; i.p.).
- QHf-AAEH: HyGR received SCF + water *ad libitum* + combined dose (ratio: 1:1:1:1 *w/w*) of *A. montanus* + *A. gangetica* + *E. coccinea* + *H. rosa sinensis* (20 mg/kg in PBS; i.p.).

Serum electrolytes

Blood volumes of 2.0 mL were drawn from 12 h post-fasted animals on the 30th day and allowed to clot in sterile sample vials. Serum samples were prepared according to the methods previously described.[25] Serum electrolytes

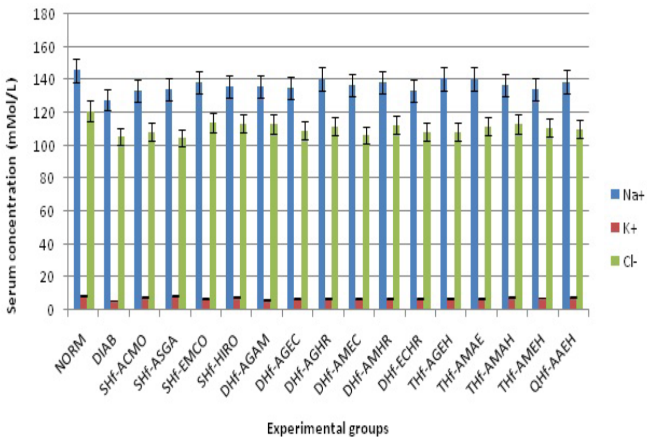

Figure 1: Serum sodium, potassium and chloride ions concentrations of normal, diabetic and treated rats

(Na^+, K^+ and Cl^-) concentrations were measured using the ion selective electrode analyzer as reported.[11]

Statistical analysis

The results were expressed as mean ± SEM, and statistically analyzed by one way ANOVA followed by Dunnett test, with level of significance set at $p<0.05$. Correlation coefficient was determined using Excel Software (Microsoft, 2010 version).

RESULTS AND DISCUSSION

Figure 1 showed that serum Na^+ concentration (SNC) of the DIAB group was significantly ($p<0.05$) lower than that of the NORM group. However, HyGR administered with SHf exhibited relatively higher SNC than that of the DIAB group ($p<0.05$), which was within the range of 133.3±13.6 mMol/L–138.2±15.3 mMol/L; $DIAB_{[Na+]}=127.8\pm12.4$ mMol/L. Likewise, SNC of HyGR administered with DHf were significantly ($p<0.05$) higher than that of the DIAB group but comparable with SNC of HyGR administered with SHf. SNC of HyGR administered with THf was not significantly different ($p>0.05$) from that of the NORM group. Specifically, $\text{THf-AGEH}_{[Na+]}=140.8 \pm 11.6$ mMol/L and $\text{THf-AGEH}_{[Na+]}=140.7\pm14.0$ mMol/L, whereas $NORM_{[Na+]}=145.9\pm11.8$ mMol/L.

Table 1: Correlation coefficients (r) of serum sodium, potassium and chloride concentrations of experimental rats.

	Na^+	K^+	Cl^-
Na^+	1.000	-	-
K^+	0.386096385	1.000	-
Cl^-	0.703231543	0.454718488	1.000

Serum K^+ concentration (SKC) of the NORM group was significantly ($p< 0.05$) higher than that of the DIAB group (Figure 1). SKC of HyGR administered with SHf was not significantly different ($p> 0.05$) from that of the NORM group; except $\text{SHf-AMCO}_{[K+]}=7.8\pm076$ mMol/L and $\text{SHf-EMCO}_{[K+]}=7.1\pm0.76$ mMol/L. Furthermore, DHf-AMEC and DHf-ECHR showed marginal improvement in SKC when compared with the DIAB group. Likewise, THf-AMAE did not exhibit significant ($p>0.05$) increase in SKC when compared with that of the DIAB group. Specifically, $\text{QHf-AAEH}_{[K+]}=8.0\pm0.11$ mMol/L was comparable with that of the $NORM_{[K+]}=9.04\pm0.13$ mMol/L; $p>0.05$.

Serum Cl^- concentration (SCC) of the DIAB group was not significantly different ($p>0.05$) from that of the SHf-ACMO and SHf-ASGA. However, $NORM_{[Cl-]}=121.0\pm13.1$ mMol/L > $DIAB_{[Cl-]} = 105.3\pm12.4$ mMol/L; $p<0.05$ (Figure 1). SCC of HyGR treated with DHf varied within the range 106.2±12.8 mMol/L–113.0±12.9 mMol/L. Also, SCC of HyGR treated with THf was not significantly different ($p>0.05$) from HyGR treated with DHf, THf and QHf.

Table 1 showed that SNC exhibited a strong positive correlation with SCC ($r=0.703231543$). Conversely, SNC versus SKC ($r=0.386096385$) and SKC versus SCC ($r=0.454718488$) gave weak positive correlations.

Aside from abnormalities of carbohydrate, protein and lipid metabolism, the present study portrays blood electrolytes levels as reliable diagnostic parameters in DM as previously described[5,6,26-29] and for monitoring therapeutic events in the course of management of DM.[30-32] Alterations in blood electrolytes is a common feature of uncontrolled DM,[5,6,13,14,31] which is exacerbated by diuretics and other medications that lowers body Na^+ levels and elicits hypovolemia.[11] In addition, compromised renal function due to prolong and uncontrolled DM engenders alteration in blood electrolytes profile by severe loss of electrolytes such as Na^+, K^+, Ca^{2+}, Cl^- and PO_4^{2-} in the urine[6,14] Afridi *et al.*, [28] proposed that deficiency of some essential trace metals may play a role in the pathogenesis and progression of DM. The strong positive correlation between SNC and SCC appeared to suggest that the metabolism of Na^+ and Cl^- are, to a large extent, intertwined and regulated in an associated manner. Also, the poor correlation ($r=0.386096385$) between SNC and SKC appeared to suggest an inverse relationship between blood levels of Na^+ and Cl- as exemplified in diabetic coma.[27]

The relatively low SNC and SKC of the DIAB group and related treated groups, compared with corresponding NORM group (Figure 1), are evidence of hyponatremia and hypokalemia in the experimental rats. Fittingly, low SNC and SCC of the DIAB group conformed to previous reports.[6,28,31] According to Wang *et al*.,[5] hyperglycemia-induced osmotic diuresis caused increased urinary excretion of Na^+, which was the underlying primary factor for low SNC in response to hyperglycemia. However, the present study showed that the various herbal formulations, in the present form, exhibited varied limited capabilities but substantial potentials to reversing altered blood electrolytes levels of HyGR within the 30-day treatment. Additionally, the positive correlation (r=0.703231543) between SNC and SCC of the experimental rats was an indication of associated regulation of Na^+/Cl^- metabolism and their definitive concentrations in plasma, could be of diagnostic relevance in the course of management and treatment DM. In furtherance to the present study, the levels of alterations of other blood electrolytes should be investigated for a broader insight into the diagnostic usefulness and significance of blood electrolytes in DM and its managements.

CONCLUSION

The present study showed that the various herbal formulations, in the present form, exhibited varied limited capabilities but substantial potentials to reversing altered blood electrolytes levels of HyGR within the 30-day treatment. Furthermore, definitive blood Na+/K+ and Cl- levels could be of diagnostic relevance in the course of management and treatment of DM.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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