Use of Phlebotomy for management of paraquat toxicity: A pilot study

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ABSTRACT

Bachground: Paraquat (PQ) intoxication manifests with pulmonary edema and fibrosis, heart failure, hepatic failure, and sometimes convulsions. In severe cases, the patients may die within the early hours. At best conditions, the conventional therapeutic methods can only save less than 50% of the victims of intentional PQ intoxication. The aim of this study was to evaluate the outcome of new therapeutic approaches particularly phlebotomy in PQ intoxication.

Methods: Five patients enrolled in the present pilot study. The conventional therapeutic method consisting of gastric lavage, diuresis techniques, corticosteroids, immuno-suppressors, and antioxidants were applied in association with new interventions including early hemo-dialysis and the use of new antioxidants, especially phlebotomy.

Results: One of the six patients who survived was excluded from the study. Four of five patients (80%) who were admitted with positive urine PQ test and treated with the modified method, could survived and only one case expired despite the interventional treatment and care. The mean age of patients was 25.21±3.47 years. Patients drank on average 52 ml of concentrated PQ. During admission, the mean Hb concentration was 16.79±2.11 g/dl that reduced to 8.95±0.93 g/dl on the second day of hospitalization.

Conclusion: Phlebotomy causes fast circulation and remove toxin subjects. This study showed effect of phelebotomy and administration of antioxidants to treat PQ-intoxication. The new modified therapeutic method with phlebotomy surprisingly increased the survival rate up to 80%.

Keywords: Herbicide, Intoxication, Management, Paraquat, Phlebotomy

INTRODUCTION

Paraquat (PQ) is a broad-spectrum herbicide which is sold under such commercial names as Gramaxon, Dextrone, and Herbaxon. It has been used since 1962 as herbicide and currently is available in more than 120 countries and is manufactured in USA, China, India, Malaysia, Brazil, and Japan (1,4). The effective component of PQ is bipyrinidium. Other toxic preparations, such as chloramquat, diquat and difenzoquat, also contain bipyridinium, but their toxic effects are lower than PQ. For instance, LD50 for diquat is 400mg/kg which is much higher than PQ LD50 (100mg /kg) (1,2).

The death picture in admitted patients is often similar to acute respiratory distress syndrome (ARDS) and follows the multi-organs failure, especially pulmonary fibrosis. However, the cause of early death is often myocardial injury, cardiac tachyarrhythmia, and shock (5). Dehydration, shock, and hypotension in early stages are due to the severe burning property of this toxin in gastrointestinal (GI) tract, leading to the secretion of liquids from burned mucosa as well as adrenal injury and the resulting secondary electrolyte disturbances (1,3). The

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cutaneous manifestations of PQ intoxication are the presence of vesicles, splitting, erythema in oral area associated with nail fissure, and sometimes nail loss or even thumb erosion (1,2).

The PQ-induced organ damage is mediated essentially with PQ monocation free radical which is produced mainly enzymatically from PQ. This intermediatory factor then promotes the well-known cascade which leads to production of reactive oxygen species (ROS), mainly hydrogen peroxide (H2O2) and hydroxyl radical (HO) (6,7).

The conventional methods can only save less than 50% of victims of intentional PQ intoxication. This has been confirmed by previous studies in USA (1,8). The prognosis in developing countries and also in Iran is even poorer. In Iran, studies have shown that the mortality rate is about 95-100% before modifying the conventional method of management (3).

The present pilot study was conducted to evaluate the efficacy of new therapeutic approach including antioxidant and phlebotomy in PQ intoxicated patients. In the present study, phlebotomy was investigated as a major part of the new approach to PQ intoxication considering the following hypotheses: a) Reducing the circulating level of PQ; b) reducing Hb level as a potential source of oxygen for ROS to a minimum level (9 g/dl); c) reducing white blood cells count as the source of ROS during organ damage; and d) triggering erythropoietin secretion as antioxidant and anti-apeoptotic agents by lowering the red blood cells count and Hb level.

MATERIALS AND METHODS

From January to August 2004, six patients were admitted to the poisoning ward of Sina Hospital of Tabriz with diagnosis of PQ intoxication. All the intoxications were due to suicidal attempts. Diagnosis of PQ intoxication was based on a history of toxin ingestion and physical examination which revealed oral ulcers, oral vesicles, and signs of chemical burning of oral mucosa which is occasionally associated with cutaneous effects of the toxin, such as nail and thumb lesions. In addition, urine PQ test was run for all of the patients (determination of plasma PQ level is not available in Iran). Package of ingested herbicide was available in four cases which confirmed the diagnosis of PQ intoxication. All the enrolled patients drank more than 30 ml of concentrated PQ.

Serum potassium (K^+) concentrations, aspartate-amino transferase (AST), and alanine-amino transferase (ALT) levels were determined, and white blood cells (WBC) count and hemoglobin (Hb) concentration measurement were conducted for all of the admitted patients during the days of admittance and the second day of hospitalization.

Conventional therapeutic method, ABCD maneuvers, gastric decontamination, urine alkalization, hemoperfusion, and administration of corticosteroids, vitamin C, and vitamin E were considered during the treatment. Also, correction of electrolyte imbalance and supportive and symptomatic care were part of our conventional treatment method.

New modified therapeutic approach, early performance of ABCD maneuvers with shortening the admission time, as well as referring the patients to intensive care unit (ICU) were done using the following procedures:

- Using gavages of Bentonite or the kind of soil which is similar to Fuller's earth dissolved in water.
- Gastric washing by concentrated solution of charcoal in water in addition to sorbitol intermittent by Fuller's earth gavages.
- Making the patient breath within a pocket and avoiding the administration of O_2 until the arterial oxygen pressure drops to under 80 mmHg.
- Urine alkalization to protect kidneys from injury and accelerate toxin excretion.
- Initiating dopamine infusion at a dose of 2-5 Micg/kg in order to resume the kidney function by using vasodilator effect of dopamine. Careful attention was paid to the maintenance of renal function in the early stages of intoxication.

- Administrating 8 mg/kg/12hrs intravenous slow infusion vitamin C and 2 IU/kg/8hrs intramuscular vitamin E, as antioxidants. Administration of vitamins was prolonged for 3-4 weeks.

- Corticosteroid therapy, administration of 200mg hydrocortisone at admission followed by methyl prednisolone succinate 250 mg every 4-6 hours.

- Administrating N-acetylcysteine (NAC) 140 mg/kg during the first 4 hrs of admission followed by 70 mg/Kg/4hrs, 17 times.

- Administrating calcium (Ca^{2+}) and magnesium (Mg^{2+}) and correction of hypomagnesaemia to prevent PQ accumulation in tissues and hypocalcaemia (calcium was administered intravenously within the first days and orally later).

- Correcting Hypokalemia by adding 30-40 mEq/lit of serum administered to the patient. The fact that pulmonary changes caused by PQ are similar to those caused by oxygen intoxicity (2) is the main motivation for using other oxygen reduction measures, such as deferroxamin that causes the reduction of O_2 transfer capacity through inhibiting the intestinal absorption of iron (we used 10mg/kg/8hrs intramuscular deferoxamine (DFO) in very ill patients).

- Utilizing symptomatic and supportive management methods which involve providing the required calories and prevention of infections.

- Inserting subclavian or femoral catheter and early HD upon admission which was repeated every 8 or 12 hours in the first day followed by diurnal HD up to one day after negative PQ test (8).

- Phlebotomy, as discussed below, as a modern method in PQ intoxication management. Phlebotomy method began with removal of 500 ml of blood. The later recommendation caused the removal of about 100 ml blood in addition to inflammatory cells precipitated within the filter wall of dialyser. The aim of phlebotomy was to lower Hb concentration to under 9 g/dl.

RESULTS

Of the six patients who survived, one was excluded from the study because of negative urine test result. From the remaining five patients who were admitted with positive urine PQ test and treated with the modified method, four could survive and only one case expired despite the interventional treatment and care. The expired patient withdrew the new therapeutic approach to PQ intoxication after two days because of his father's disagreement. The mean age of patients was 25.21±3.47 years. Patients drank 52 ml of concentrated PQ (ranged 30-65 ml) on average. Mean WBC count was 8132.00±1022.80 m^3 at the time of admission and 15920.00±2804.81 m^3 on the second day of hospitalization in spite of phlebotomy. The mean Hb concentration was 16.79±2.11 g/dl upon admission that decreased to 8.95±0.93 g/dl on the second day of hospitalization. Mean K^+ levels were, respectively, 3.86±1.06 and 3.74±0.55 on the first and the second days despite electrolyte corrective treatments.

At the time of admission, AST and ALT levels were 25.18±4.56 and 19.74±3.80, respectively, while on the second day of hospitalization, they reached 29.66±3.61 and 22.41±5.73, respectively. Among the five patients with PQ intoxication, four finally survived without any remission after a five-year follow up.

DISSCUSSION

PQ acts as herbicide through interaction with intracellular electron transport system and inhibition of reduction of NADP to NADPH along with photosynthesis (9). After ingestion, PQ leads to the production of superoxide, peroxide, and hydroxyl radicals. In addition to superoxide and hydroxyl radicals which are produced by bipyridinum toxicity, metal ions are also developed in human tissues due to biochemical stimulation of PQ which accelerates the production of lipid peroxides (1, 2, 10, 11). After ingestion of about 35 mg/kg or 50 ml of concentrated PQ (with LD50 of 100mg/kg), the patient presents systemic intoxication manifestations including pulmonary edema, heart failure, hepatic failure, and sometimes convulsions

(acute intoxication). Mild intoxication may occur by lower amounts of PQ as much as 4 mg/kg (1, 2, 11).

The effect of PQ on respiratory tract is manifested by dizziness, excessive saliva, cough, respiratory distress, epistaxis, and hemoptysia which progress to pulmonary edema and extensive fibrosis. The onset of respiratory symptoms and respiratory distress may be delayed for several days (5). Regarding the stimulatory effect of this toxin on collagen synthesis and its pulmonary accumulation as well as the presence of maximum oxygen content in the lungs, the lungs are considered appropriate sites for manifestation of symptoms and development of extensive and progressive fibrosis (2). The cause of fibrosis in this case is inhibition of superoxide dismutase (SOD) leading to stimulation of inflammatory cells. The stimulation of inflammatory cells in PQ intoxication is a well-defined process, but elevation of leukocyte count in proportion to the severity of intoxication as well as partial elevation of red blood cells (RBC) count was noted newly in our experiences. The elevation of neutrophil/lymphocyte ratio to 80-90%/10-20% was another new finding in our blood analysis in patients with PQ intoxication.

Three important factors are involved in pathophysiology of PQ intoxication consisting of oxygen (because of its free radicals), inflammatory reactions and cells (especially neutrophils), and the PQ compound itself. Moreover, PQ stimulates fibroblasts and collagen synthesis (1, 3, 5). Conventional methods include removing PQ from the gastrointestinal tract, increasing its excretion from blood, and preventing pulmonary damage with anti-inflammatory agents. For this reason, gastric decontamination is performed routinely by bentonite and Fuller's earth. Bentonite can be poured only in a 6 to 8 percent suspension and should be administered if Fuller's earth is not available. Fuller's earth can be administered in a 30% suspension at which it remains pourable along with magnesium sulfate.

Diuresis techniques are also applied to intoxicated patients but they are likely to be effective within only the first 24 hours after ingestion. Hemoperfusion and hemodialysis have been suggested as measures for removing PQ from blood (1, 3, 5).

Corticosteroids have traditionally been used for PQ intoxication although their beneficial effects are in doubt. In addition, other agents including azathioprine, beclomethasone, and bleomycin have been used without their clear-cut benefits being shown. Oxygen administration to patients appears to accelerate the lung diseases and it is suggested that an inspired oxygen concentration of greater than 21% should be used only when arterial oxygen pressure drops to below 80 mmHg.

In addition, the combined use of antioxidants such as DFO, NAC, vitamin C and E was proposed in the case of PQ intoxication (1, 3, 5, 12). It has been shown that DFO can exert its protective effects, not only by inhibiting the PQ-induced generation of hydroxyl radicals, but also by blocking the uptake of PQ by the alveolar type II cells (13). Exposure of human alveolar cells in-vitro to PQ produced apoptotic cells death, perhaps via oxidative stress mechanisms. This toxic effect was inhibited by NAC, a treatment effect attributed to the direct scavenging action of the sulphydryl group of NAC (14). The mechanism for such a protective effect of vitamin C has been attributed to its ability to quench radicals generated by the redox cycling of PQ before they attack other biomolecules. Ascorbic acid acts as a two-electron reducing agent and confers protection by contributing an electron to reduce free radicals, thus neutralizing these compounds in the extracellular aqueous environment prior to their reaction with biological molecules (15, 16). High concentrations of ascorbic acid are found naturally in lung fluids to protect it against free radicals generated by toxic chemicals in air, such as ozone, sulfur dioxide, metal fumes, and cigarette smoke (15, 17). Moreover, the antioxidant potential of ascorbic acid is not only attributed to its ability to quench reactive oxygen species, but also to its ability to regenerate other small molecule antioxidants, such as a-tocopherol, glutathione, and b-carotene (15, 16, 18). Vitamin E is a lipid-soluble vitamin that exerts

its antioxidant effects by scavenging free radicals and stabilizing membranes containing polyunsaturated fatty acids (19, 20). Results from in-vivo studies have demonstrated that vitamin E administered in large doses over a prolonged period of time confers protection against oxidant-induced tissue damage (21-24). Some animal studies have suggested that other uncommon antioxidants such as 3-methyl-1-phenyl-2-Pyrazolin-5-one are able to reduce the adverse effects of PQ (25).

Our study of blood samples obtained from the victims of PQ intoxication showed that

PQ causes the elevation of WBC count, especially neutrophils, during the first hours of intoxication. It appears that initial phlebotomy at 500 ml within the first hours, repeated every 12 hours as well as before every dialysis is effective. Phlebotomy had the following advantages:

- Phlebotomy mainly (particularly primary fasted 500 ml blood) helped remove the

circulating toxin faster, especially within the earlier hours of intoxication which are

critical.

- Phlebotomy removed the neutrophils which occasionally constitute 80-90% of all WBC. Surprisingly, WBC count reached 13000-17000/µl in the studied patients and even 25000/µl in severe cases. PQ causes a significant increase in the total number of WBCs (26). An early feature of PQ toxicity is the influx of inflammatory cells, releasing proteolytic enzymes and ROS, which can destroy the lung epithelium and result in pulmonary fibrosis. Neutrophils exposure to PQ was also used to increase intracellular O2 levels (27, 32). These experiments demonstrated that increased intracellular O2 induces proi-nflammatory cytokine production in neutrophils and further its protrude role in increased ROS production. The epidemiologic study by Lee et al (33) reported a good prognostic factor of survival by lesser degrees of leukocytosis after acute paraquat poisoning. Considering neutrophil count decrease and total WBC together may be important and helpful. In addition, phlebotomy reduces other inflammatory cells leading to a significant reduction in pulmonary fibrosis

- Hb concentration reaches up to 16 g/dl during PQ intoxication. Studies investigating the predictors of survival in PQ intoxicated patients showed that higher levels of Hb are associated with worsened prognosis (33). Reducing this rate to 9 g/dl results in reduced oxygenation of body viscera, preventing further injury by toxin-oxygen complex and free oxygen radicals in combination with inflammatory cells and neutrophils.

Major limitations of this study could be the very small sample size, the absence of a control group, and the limited number of assessed biochemical parameters (especially shortage of plasma PQ level, erythropoietin level, total antioxidant capacity, and ROS concentration). To the best of our knowledge, it is the first study attempting to show the effect of phlebotomy and new antioxidants on PQ intoxication. Further (experimental and clinical trial) studies with larger sample sizes need to be conducted to determine the role of phlebotomy in ROS, antioxidants enzymes, and outcome of PQ intoxicated patients.

In the present study, combinational use of dopamine, DFO, and phlebotomy along with other conventional measures resulted in complete cure of four PQ intoxicated patients (80% cure and survival).

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