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Vanadium-Induced Leukocytosis

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Vanadium is a micronutrient (Hopkins and Mohr 1974) and shown to be an important element in carbohydrate metabolism (Korovkin et al 1995), as well as in other key metabolic events (Chasteen 1983). A progressive increase in the soil concentrations of vanadium has been reported (Biernacka et al 1983). Elevated vanadium concentrations appear in the environment due primarily to its mobilization by fuel combustion and its use in a number of industrial processes (Meneses et al. 1999 and Abbasi 1987). As a consequence. increased vanadium levels have appeared in plant species. This, of course. increases the vanadium consumption and retention in recipient species (Welch and Cary 1975) promoting vanadium toxicity. As a result of the increased level of vanadium appearing in animals, more experimental attention has been directed to the study of the biotoxic effects of vanadium.

At above the estimated essential micronutrient level of vanadium, a broad spectrum of pathophysiological states have been reported. These include growth pattern modification (Schwarz and Milne 1971), a variety of renal effects (Hogan 1989), and respiratory (Knecht et al 1985) and cardiovascular system dysfunction (Borchard et al 1981). The effects ofvanadium are known to be variable depending upon a number of factors including the oxidation state of the test compound. In regard to this variable, vanadyl sulfate [V(IV)] was reported to be more effective in altering renal function (Hogan 1989) and inducing hemolysis (Hogan 1990) in rodents compared to the effects of a trivalent and pentavalent vanadium compounds.

The primary objective of the report published here was to explore further the toxicity of vanadyl sulfate on peripheral blood. Specifically, the study examines vanadyl effects on the level and class of circulatory leukocytes of mice at selected times following a single treatment of the test compound.

METHODS AND MATERIALS

Laboratory-reared young adult female mice of the Institute of Cancer Research (ICR) strain were used throughout these investigations. Ten mice comprised each experimental and control subgroup. Animal weights ranged between 26 and 33 g with an average of 27.8. On d 0 vanadyl sulfate (Aldrich Chemical Co., Inc.) solution was prepared at pH 7.4 and injected (intraperitoneal) at a 2.0 mg/kg dosage. Control mice received 0.3 ml isotonic saline which represented the average injectate volume for the vanadium-treated animals.

On d 1, 3, 5, 7 and 9, ten experimental and control mice were sacrificed by cervical dislocation after which cardiac blood was withdrawn using sodium heparin as the anticoagulant. From the blood sample, the total leukocyte count (wbc/mm3) for each sample was determined using a Coulter T-540 Hematology System. Blood smears were prepared on pre-cleaned microscope slides followed by standard hematological staining with Wrights-Giemsa (Wintrobe 1956). Differential leukocyte counts were determined from these slides using the cytological criteria according to Bessis (1977). After differential scoring, the percentage of granulocytes (neutrophils, basophils, and eosinophils) was compared to that of agranulocytes (lymphocytes and monocytes) at the various sampling intervals following the d 0 vanadyl sulfate injection. Data were analyzed for their statistical significance using the Analysis of Variance.

RESULTS AND DISCUSSION

The studies presented here report that a single intraperitoneal injection of vanadyl sulfate induced an abrupt increase in the number of circulating white blood cells or leukocytosis beginning one day after injection and continued to d 7 (Figure 1). On d 1 following vanadium, the leukocyte count increased to approximately 5,200 wbc/mm^3 compared to the control value of about 4,100 wbc/mm^3 or a 26.8 percentage increase. This trend continues on d 3. 5 and 7 for increased circulating leukocyte percentage of 27.3, 40.5, and 20.9. respectively. These values are statistically different ($p<0.01$ for days 1 and 7 and $p<0.001$ for days 3 and 5) from those of saline-injected control female mice. On d 9, the leukocyte counts determined from saline- and vanadium-injected mice were not statistically separable.

The leukocytosis following vanadium injection was due primarily to an elevation in the number of circulating granulocytes (Fig. 2). It is noted that on d 1 vanadium-treated animals had an approximately 10 higher percent points

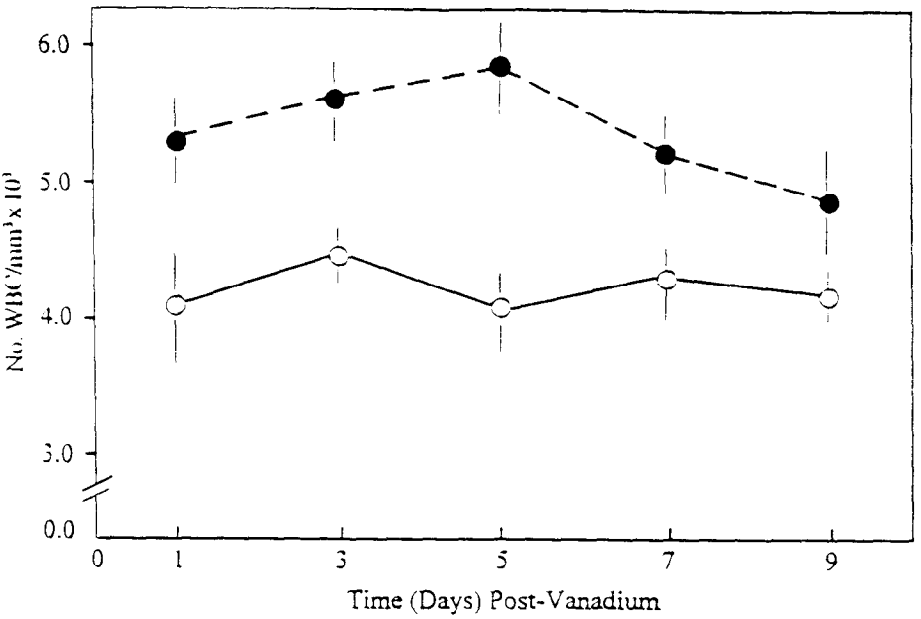

Figure 1. Time course of leukocyte response to vanadium treatment (closed circles) administered on day 0 compared to saline-injected controls (open circles). Each point represents the mean count obtained from 10 animals. Vertical lines are the standard errors of the means.

than controls and the difference continued to d 3 (16.5 percent) and d 5 (21 percent). By d 7 and 9 the granulocyte percentages of the experimental and control animals did not differ. For agranulocytes, the percentage decreased for vanadium-injected mice was a mirror image of the granulocyte increase. From the differential counts, if the number of granulocytes increased, the probability of scoring granulocytes would be greater. Therefore, even if the agranulocyte level did not change in the absolute sense, relatively speaking, the probability of "seeing" these cells would by less due to the increased number of granulocytes observed during differential scoring. This would account for the elevated white blood cell count due primarily to a granulocytosis. Others have reported similar results, i.e., an increase in the number of white blood cells and relative percentages of granulocytes in monkeys inhaling vanadium pentoxide (Knecht et al 1985).

The data of the current study are in contrast to observations by Dai et al. (1995) who orally administered vanadyl sulfate to rats and reported no demonstrable effect on erythrocytes, leukocyte or platelet counts. Similarly,

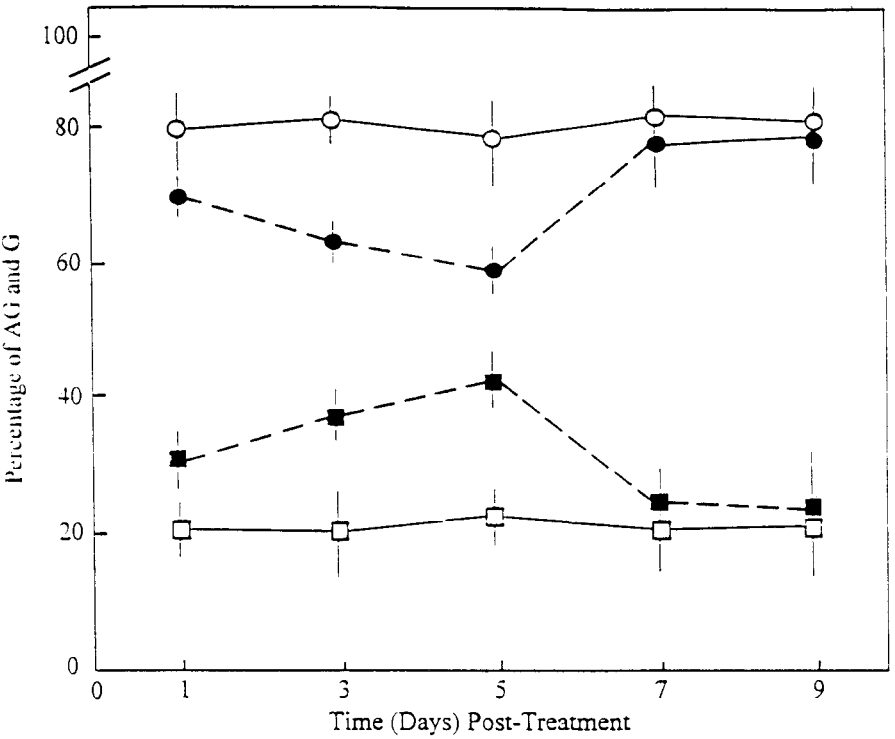

Figure 2. The percentage of agranulocytes (Ag, closed circles) and agranulocytes (A, closed squares) of vanadium-treated mice and saline-injected controls (open circles and squares). Vertical lines of points represent standard errors of the means.

Fawcell et al. (1997) found no hematological changes following orally delivered vanadium to humans. The obvious difference in those studies and the one reported here is the route of administration of vanadium in addition to the test species variable. A vanadium effect on leukocytes has been reported (Zaporowska and Wasilewski 1990) in rats receiving vanadium in their drinking water: this work reported a rise in the leukocyte counts due primarily to an agranulocyte increase (particularly lymphocytes). In Figure 3 the differential percentages of agranulocytes and granulocytes do not differ on d 7 from control percentages. This indicates that at that time both classes of leukocytes had increased proportionately to control values during the increase in the absolute number of wbc/mm^3. The later increase for the agranulocytes could be due to a stimulating production response. It is tempting to postulate that after the intraperitoneal infection, the vanadium enters the circulation to cause tissue trauma and inflammation. This would in turn promote migration of granulocytes, especially the highly chemotaxic neutrophils, from reserve tissue sites into the blood causing a leukocytosis. Because of the early response that was observed, this notion seems more attractive than one based

upon the granulocyte elevation due to an increased bone marrow production.

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