Mycobacterium Cell Wall Fraction (MCWF) as an aid in the treatment of chemotherapy-induced neutropenia in dogs

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Chemotherapy-induced neutropenia in clinical oncology may have negative implications as it increases the risk of infection, interrupts chemotherapy protocols and requires treatment dose reduction. Currently, there is no veterinary product available for use in the prevention of chemotherapy-induced neutropenia. For many years immune response modifiers have been used for cancer treatment either alone or in conjunction with standard treatment protocols including surgery, radiation or chemotherapy. Mycobacterium Cell Wall Fraction (MCWF) is a non-specific immunomodulator derived from a non-pathogenic mycobacterium, *Mycobacterium phlei* which has been used for treatment of mammary tumors, osteosarcoma, transitional cell carcinoma, hemangiosarcoma and transitional venereal tumors in dogs. In addition, MCWF is currently used in Phase III clinical studies in humans for patients with bladder cancer who failed standard treatment protocols (BCG). Main purpose of our research was to demonstrate MCWF immunomodulatory potential in ameliorating the chemotherapy-induced neutropenia in dogs.

Efficacy of three different doses of MCWF as an aid in the treatment of neutropenia induced by chemotherapeutic agent vinblastine in healthy dogs was evaluated. Study design included four experimental groups with ten dogs per group (1-8 years of age). Dogs were required to be healthy to proceed to vinblastine treatment for induction of neutropenia (defined as Day 0), and further, were required to demonstrate neutropenia within 7 days of vinblastine treatment, to proceed to inclusion in the study, and treatment with MCWF. Neutropenia was defined as less than 2,000 neutrophils per µL of blood for the purpose of this study. All dogs received 3 mg/m^2 of vinblastine on day 0, by the intravenous route (IV), to induce neutropenia, then, once neutropenia was attained, a dog was randomly allocated to one of the 4 treatment groups (100, 200 or 500 µg/kg MCWF IV, or no treatment). MCWF treatment was administered by IV route as a slow bolus injection (3-5 min.) within 24 hours. CBC counts were measured on Day 0, Day 2 and daily until the end of the study. The end of the study was defined as neutrophil count ≥ 2000/µL for two consecutive days (48 hours) or day 10, whichever came first. CBC counts included monitoring of packed cell volume (PVC), red blood cells (RBC), thrombocytes, total white blood cells (WBC), and differential WBC (neutrophils, lymphocytes, monocytes, eosinophils and basophils). Efficacy of MCWF as a treatment for neutropenia was determined by comparing the duration of neutropenia in the control group versus the three different MCWF dose groups. Duration is defined as the number of days from MCWF treatment administration (24 hours after neutropenia was detected) until neutropenia was resolved (neutrophil count ≥ 2000 cells/ µL).

A clear dose-response was apparent in the time to recovery from neutropenia; mean duration of neutropenia was 4.2, 2.5, 1.8, and 1.2 days in the control, 100, 200 and 500µg/kg MCWF-treated groups respectively (Table 1.). The difference in the mean duration of neutropenia between the control and 100 µg/kg MCWF-treated groups was statistically-significant at $p<0.05$, the 200 and 500 µg/kg MCWF-treated groups also had a mean duration of neutropenia significantly shorter than the control group, with $p<0.01$ and $p<0.0001$ respectively (Figure 1). The differences in mean recovery time between the 100 and 200 µg/kg dose groups ($p<0.05$), and the 200 and 500 µg/kg dose groups ($p<0.05$) were also statistically-significant. Dogs were observed for safety of administration of MCWF, for 30 minutes immediately after administration, and again at 12 hours after administration. Adverse events such as dizziness, lethargy, and mild transient hyperthermia were observed in some dogs immediately after MCWF administration but resolved within 12-24 hours and did not require additional treatment. Vomiting and diarrhea were observed to a lesser extent as expected. All GIT adverse events were mild and lasted for 2 to 3 days following vinblastine treatment (strictly chemotherapy associated).

Administration of MCWF demonstrated efficacy in ameliorating chemotherapy-induced neutropenia in healthy dogs. These findings could have significant importance from a clinical standpoint and could support the use of MCWF in conjunction with standard chemotherapy protocols. Additional studies are underway to demonstrate the efficacy of MCWF in preventing chemotherapy-induced neutropenia following concurrent administration in tumor bearing dogs.

Table .1. Median and mean neutropenia grades by treatment group

Treatment group	Neutropenia grade (median value)	Median duration (days)	Neutropenia grade (mean value)	Mean duration (days)
Control	4	4	3.7	4.2
100 µg/kg	3.5	2	3.3	2.5
200 µg/kg	3	2	3.2	1.8
500 µg/kg	4	1	3.6	1.2

Severity grading system: 1 1,500 – 1,999 neutrophils/µL; 2 1,000 – 1,499 neutrophils/µL; 3 500 – 999 neutrophils/µL; 4 < 500 neutrophils/µL

Figure1. Time-course of neutropenia development and resolution following vinblastine administration.