VITAMIN C BUT NOT VITAMIN B2 AMELIORATES MUCOSITIS SYMPTOMS IN THE METHOTREXATE-INDUCED MUCOSITIS RAT MODEL

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Introduction

- Mucositis is a side-effect of chemotherapy treatment. In the gut, mucositis is associated with symptoms such as diarrhoea, pain, neutropenia, bacteraemia and malnutrition [1].
- Chemotherapy drastically alters the diversity and composition of the intestinal microbiota. A decrease in commensal bacteria is regularly reported in patients undergoing chemotherapy, coupled with an increase in a number of bacterial subtypes [2].
- Redox active compounds, such as vitamins B2 and C, were shown to reduce inflammation in the gut and to enhance the growth of anaerobic bacteria [3].

Aim

This study aims to (i) evaluate impact of vitamin B2 and C supplementation on growth characteristics of anaerobic bacteria under oxidative conditions, and (ii) to determine the prophylactic efficacy of vitamin B2 and C on methotrexate-induced mucositis using a validated rat model

Materials and methods

Assessing bacterial growth

To investigate the effects of vitamins B2 and C, alone or in conjugation, on the growth of six bacterial strains isolated from MTX-induced mucositis rats, their growth medium was enforced with different concentrations of these vitamins and the optimal density measured for 48 hours using Biotek.

Methotrexate-induced mucositis rat model [4]

MTX-induced mucositis rats were intravenously injected with MTX (45 mg/Kg) or saline solution (control). Vitamins B2 and C were daily administrated via oral gavage, starting one day before MTX treatment. Mucositis severity was determined by food intake, body weight and water intake. Blood was collected at days 0, 2, 4, 6 and 10 to measure plasma citrulline levels.

Figure 1. Schematic representation of the experimental design used for the *in vivo* study.

Results

Vitamins B2 and C improved the growth of anaerobic bacteria under oxidative stress

Figure 2. Growth curves of bacteria isolated from MTX-induced mucositis rats stimulated with Vitamins B2 and C. *E. coli*, *B. coccoides*, *R. intestinalis* and *E. faecalis* were grown in yeast extract, casitone, fatty acid and glucose medium and vitamins were added to the medium. Experiment was performed under oxidative conditions.

Different doses of vitamin B2 failed to improve mucositis symptoms

Figure 4. Relative body weight, food intake, water intake and plasma citrulline levels. Vitamin B2 (10, 30 and 60mg/kg/day) was administrated via oral gavage. (A) Body weight relative to day 0 was set to 100%. (B,C) Food intake and water intake in gram per day. (D) Plasma citrulline levels in μmol/L at days 0, 2, 4, 6 and 10 after MTX injection.

Vitamin C ameliorates mucositis symptoms in rats

Figure 3. Relative body weight, food intake, water intake and plasma citrulline levels. Vitamin C (250mg/Kg/day) was administrated every day via oral gavage. (A) Body weight relative to day 0 was set to 100%. (B,C) Food intake and water intake in gram/ml per day. (D) Plasma citrulline levels in μmol/L at days 0, 2, 4, 6 and 10 after MTX injection.

Conclusions

- Anaerobic bacteria *Blautia coccoides* and *Roseburia intestinalis* significantly grow better in the presence of oxygen, when a combination of 0.5 mM of both vitamins is added to the culture.
- Vitamin C supplementation (250mg/Kg/day) in rats attenuated the severity of mucositis reflected by changes in body weight, food intake and plasma citrulline.
- 10, 30 and 60 mg/Kg/day of vitamin B2 were found to have a negative impact on mucositis, suggesting that vitamin B2 worsens mucositis.

References

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