Iodide reduces cachexia in a BALB/c CT26 mouse tumor model

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Introduction: Faraday Pharmaceuticals is focused on the research and development of elemental reducing agents (ERAs) and their potential applications for the treatment of critical care diseases with a focus on cardiac and skeletal muscle disease. In this study we investigated the utility of FDY-5301 (sodium iodide) or Bucindolol (as a positive control) in reducing cachexia in a CT26 mouse tumor model.

Methods: Male Balb/c mice were injected subcutaneously with a CT26 tumor cell line. Once the tumor volume reached ~125 mm³ the animals were randomized into four treatment groups: vehicle control, 2 mg/kg/day FDY-5301 i.v., 2 mg/kg/day Bucindolol p.o., and 40 µg/day FDY-5301 delivered continuously by subcutaneous osmotic pump. Clinical signs, body weight and food consumption were monitored daily, and tumor volume was monitored every 3 days. A subset of animals was sacrificed on day 14 for plasma iodide analysis. At the end of the experimental period (day 20) the remaining animals were sacrificed, and blood was collected for biochemical analysis of glucose, total protein, a lipid panel, and cytokine analysis. Tissues and organs were weighed and fixed. Heart, lung, spleen, kidney, and epididymal fat. The muscles: gastrocnemius, tibialis anterior, and soleus, were weighed and then fixed, H&E stained and analyzed morphometrically.

Results: Administration of FDY-5301 and Bucindolol significantly increased overall body weight, liver, heart, and tibialis anterior muscle weight compared to vehicle treatment, and significantly decreased tumor growth. The group receiving continuous delivery of FDY-5301 showed a significant increase in tibialis anterior muscle fiber area compared to vehicle control. FDY-5301 (i.v.) and Bucindolol helped preserve epididymal fat weight, while FDY-5301 (continuous delivery) and Bucindolol stemmed the rise in triglycerides and VLDL.

Conclusions: Administration of FDY-5301 and Bucindolol helped retain the body weight of animals, prevented organ and skeletal muscle cachexia, and reduced tumor growth.

Materials & Methods

• Male Balb/c mice (~7–8 weeks old) from Environ, were group housed in individually ventilated cages (IVC) with 15-20 fresh air changes per hour and maintained in a controlled environment with a temperature of 22 ± 2 °C, humidity of 50 ± 10%, and a 12 hour light/dark cycle.
• Autochleaved cornflakes (Spaeroon Life Sciences, Bangalore, India) was used as a bedding material. The animals were fed ad libitum, with certified irradiated laboratory rodent diet (Nutritional Brand, Tetrion Chemical Pvt. Ltd., Bangalore) and had access to fresh, RO filtered water, during the study period.
• CT26 (murine colon carcinoma) cells with a viability of >90% were injected subcutaneously into the right flank of male Balb/c mice (~1x10⁶ cells/vial) and maintained for tumor growth. Once the tumor volume reached ~125 mm³ the animals were randomized into groups.
• Group I - non-tumor bearing healthy control
• Group II - vehicle control, p.o. administration of 0.5% carboxymethyl cellulose (CMC)
• Group III – FDY-5301 i.v, 2 mg/kg/day (saline as the vehicle)
• Group IV – Bucindolol p.o., 2 mg/kg/day (0.5% CMC as the vehicle)
• Group V – FDY-5301, 40 µg/day delivered via Alzet slow release osmotic pump.
• The Alzet® pump, model 1004, from DURECT (Cupertino, CA) was filled with FDY-5301 and soaked overnight in saline to prime prior to sterile implantation into the left flank of the day on randomization.
• Clinical signs, body weight and food consumption were monitored daily. Tumor volume was monitored every 3 days using a Vernier caliper to determine length (L) and width (W). TV (mm³) = (L x W²/2).
• A subset of animals were sacrificed on day 14 for plasma iodide analysis. Iodide concentration was determined using ion chromatography with an amperometric detection.
• At the end of the experimental period (day 20) the remaining animals were sacrificed, and blood was collected for biochemical analysis of glucose, total protein, a lipid panel, and cytokine analysis. Various tissues and organs were weighed including: tumor, liver, heart, lung, spleen, kidney, and epididymal fat.
• The muscles: gastrocnemius, tibialis anterior, and soleus, were weighed and then fixed, H&E stained and analyzed morphometrically.
• For the evaluation of the statistical significance of organ and food weight two-way ANOVA and for tumor growth inhibition two-way ANOVA followed by Bonferroni post hoc test was performed using Graph Pad Prism v5.0. p values <0.05 indicate statistically significant differences between groups.
• Animals were taken care as per the regulations of Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), of Government of India and Association for Assessment and Accreditation of Laboratory Animal Care (AAALAC) guidelines.

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