

Interaction of dietary *Bacillus subtilis* and fructooligosaccharide on the growth performance, non-specific immunity of sea cucumber, *Apostichopus japonicus*.

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Interaction of dietary *Bacillus subtilis* and fructooligosaccharide on the growth performance, non-specific immunity of sea cucumber, *Apostichopus japonicus*

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### Abstract

A feeding experiment was conducted to investigate the interaction of probiotic *Bacillus subtilis* and prebiotic fructooligosaccharide (FOS) on the growth performance, immunity, intestinal microflora and disease resistance of sea cucumber (*Apostichopus japonicus*). Five hundred and forty individuals (initial body weight:  $5.06 \pm 0.10$  g, mean  $\pm$  S.E) were fed nine practical diets according to a  $3 \times 3$  factorial design: the basal diet as the control diet supplemented with three levels of *B. subtilis* (0,  $1.82 \times 10^7$  or  $4.95 \times 10^7$  CFU  $g^{-1}$  diet), crossed with 0, 0.25% or 0.50% FOS. After 8 weeks, three sea cucumbers per tank were sampled for bacterial

quantification and immunity determination. Then all the sea cucumbers left were challenged by *Vibrio splendidus*. The results showed that dietary *B. subtilis* significantly increased the specific growth rate (SGR), total coelomocytes counts (TCC), phagocytosis of sea cucumbers, the counts of total viable bacteria and disease resistance to *V. splendidus* ( $P < 0.05$ ), whereas the counts of *Vibrio* decreased. However, dietary *B. subtilis* had no significant effect on phenoloxidase (PO) activity in coelomocyte lysate supernatant (CLS) ( $P > 0.05$ ). The SGR, PO activity, total viable bacterial counts (TBC) and *Vibrio* counts (VBC) were significantly affected by dietary FOS. In the group with 0.50% FOS, TCC, phagocytosis and PO activity significantly increased compared to the group fed without FOS in diet ( $P < 0.05$ ). In the groups with  $1.82 \times 10^7$  CFU *B. subtilis*/g diet, FOS supplementation remarkably decreased VBC. And higher level of FOS (0.50%) resulted in significantly higher TCC and PO activity compared with 0.25% FOS ( $P < 0.05$ ). Moreover, the animals fed with diets supplemented with 0.25% and 0.50% FOS at each *B. subtilis* level had notably lower cumulative mortality after 14 days following *V. splendidus* exposure ( $P < 0.05$ ). Under the experimental conditions, dietary *B. subtilis* and FOS had a synergistic effect on enhancing immunity and disease resistance of sea cucumber ( $P < 0.05$ ).



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## Keywords

*Apostichopus japonicus*; Probiotic; Prebiotic; Growth; Immunity; *Vibrio splendidus*

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