Povidone iodine exposure alters the immune response and surface microbiota of koi carp, *Cyprinus carpio*.

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Abstract

We aimed to assess the effects of Povidone-iodine (PVP-I) on the immune system and bacterial communities of skin and gill in *Cyprinus carpio*. Fish were disinfected with PVP-I for 30 min, and then skin and gill tissues were sampled on days 0, 1, 3, 7, and 14 for the determination of immunity parameters and characterization of surface bacterial communities. The results indicated PVP-I decreased the activities of gill lysozyme on day 1, and significantly increased the activities of gill SOD and Na-ATPase on day 7. Skin SOD activity was enhanced on day 3. The PVP-I bath enhanced the mRNA expression of *lysozyme, sod,* and *cat* in the gill but decreased *lysozyme* on days 1-7 and *sod* on days 7-14 in the skin. The mRNA expression of tight junction proteins enhanced in both gill and skin tissue. PVP-I decreased gill bacterial richness on days 3 and 7, and reduced bacterial diversity on day 7. The gill microbiota on day 7 was remarkably separately from the control. The skin microbiota on days 1 and 3 were significantly separated from that of the control. In summary, PVP-I elicited shifts in the immune response and changes in the surface microbiota of fish.

Methods

- A total of 54 fish were disinfected with 7.5 mL/m^3 of PVP-I (1%) for 30 min. Fish (n = 9 per treatment) were sampled at six time points: one hour before PVP-I treatment (C0d), and then at 0h (immediately after PVP-I disinfection, T0d), 1d (T1d), 3d (T3d), 7d (T7d), and 14d (T14 d) after the PVP-I treatment shown in Figure 1.
- All fish were immediately anaesthetized with tricaine methanesulphonate (MS-222). Each fish was then flushed with approximately 500 mL of germ-free water. Skin samples were carefully removed from the left side of the fish body from the area behind the operculum and above the lateral line, and then stored in a sterilized centrifuge tube. The first left gill arch was excised and collected in another sterilized centrifuge tube. All samples were stored at −80°C for further measurement of enzyme activities, antibodies, and bacterial community composition.

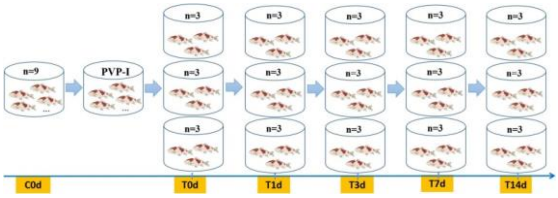

Fig. 1 Schematic figure of the experimental design.
C0d: Fish were sampled prior to PVP-I disinfection; T0d: Fish were sampled immediately after PVP-I disinfection; T1d: Fish were sampled on day 1 after PVP-I disinfection; T3d: Fish were sampled on day 3 after PVP-I disinfection; T7d: Fish were sampled on day 7 after PVP-I disinfection; T14d: Fish were sampled on day 14 after PVP-I disinfection.

Results

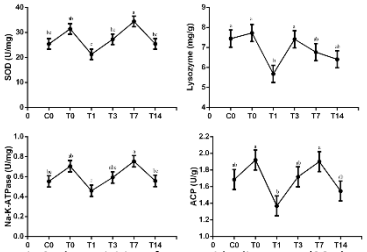

Fig 2: The activities of superoxide dismutase (SOD), lysozyme, Na/K-ATPase, and acid phosphatase (ACP) in gill tissue.

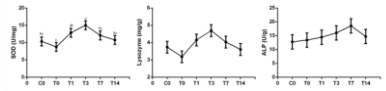

Fig 4: The activities of superoxide dismutase (SOD), lysozyme, and alkaline phosphatase (ALP) in skin.

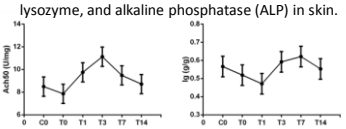

Fig 5: The concentration of alternative complement activity (ACH50) and Total immunology (Ig) in skin.

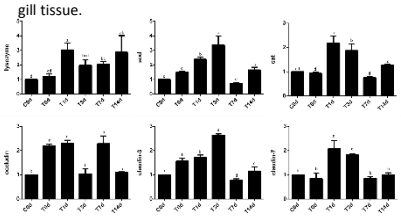

Fig 3: Relative expression of lysozyme, sod, cat, occludin, claudin-3, and claudin-7 genes in the gills of fish treated by PVP-I.

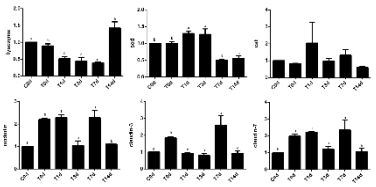

Fig 6: Relative expression levels of lysozyme, sod, cat, occludin, claudin-3 and claudin-7 genes in the skin treated by PVP-I.

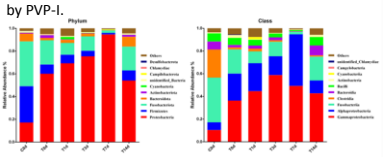

Fig 7: The average relative abundance of top 10 most abundant bacteria of gill tissue at the (A) phylum and (B) class level.

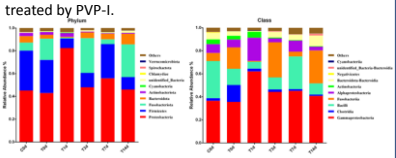

Fig 10: The relative abundance of skin bacteria at the (A) phylum and (B) class level.

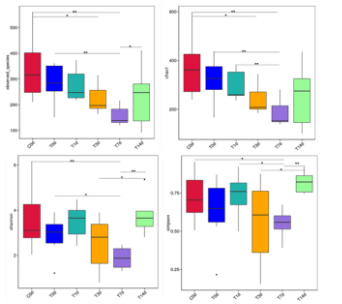

Fig 8: The Observed species, Chao1, Shannon, and Simpson indices of gill bacteria.

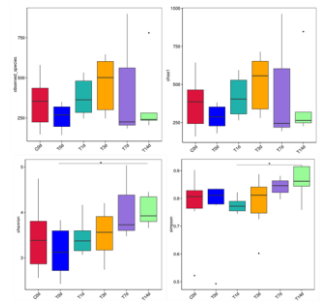

Fig 11: Observed species, Chao1, Shannon, and Simpson indices of skin bacteria.

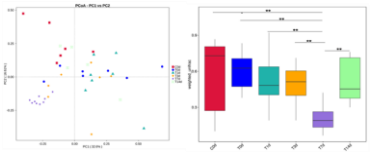

Fig 9: Principal coordinate analysis of the gill bacterial community based on weighted UniFrac distance.

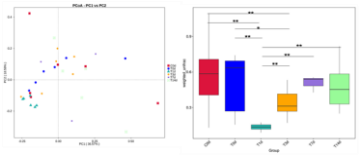

Fig 12: Principal coordinate analysis of skin bacterial community.

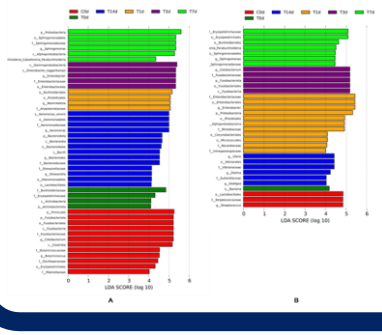

Fig 13: Taxonomic comparisons of the bacterial communities among treatments by LEfSe

Conclusion

In summary, PVP-I elicited shifts in the immune response and changes in the surface microbiota of fish.

Acknowledgments

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