Short Paper

The Vitamin B_{12}-Producing Ability of Intestinal Bacteria Isolated from Tilapia and Channel Catfish

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Lovell and his co-worker[1,2] reported that the apparent rate of intestinal bacterial synthesis of vitamin B_{12} in tilapia *Tilapia nilotica* fed a vitamin B_{12}-deficient diet (at least 11.2 ng per g of body weight per day) was eight times that found in channel catfish *Ictalurus punctatus* fed the same diet (ca. 1.4 ng/g/day) although they did not compare the intestinal microflora of those fish species. There is little information on the vitamin B_{12}-producing ability of fish intestinal bacteria except for carp *Cyprinus carpio*.[3] The present study was carried out to investigate the vitamin B_{12}-producing ability of intestinal microflora from tilapia and channel catfish.

Feces were removed from each of the four specimens of tilapia (18.9–45.8 g in body weight) and channel catfish (3.9–5.3 g) which had been fed a pelleted diet (Nippon Haigo Shiryo), and microbiological examination was performed according to the previous paper.[4] About three strains for each genus which showed the highest count through the seven agar media used, were selected from each specimen and incubated in GAM broth (Nissui) at 25°C for 5 days under aerobic or anaerobic conditions. The culture was then assayed microbiologically for vitamin B_{12} using *Lactobacillus leichmannii* IAM 12066 (=ATCC 7830),[5] and the amount of vitamin B_{12} produced or consumed was calculated from the difference between the inoculated and uninoculated broths.

Viable count (CFU/g) of each bacterial genus in the tlapia's intestine was: *Aeromonas*, 2.5×10^6–3.0×10^7; *Plesiomonas*, 2.0×10^6–7.7×10^7; *Enterobacteriaceae*, nd (not detected)–1.8×10^7; *Pseudomonas*, 4.5×10^6–5.6×10^9; *Moraxella*, nd-2.8×10^4; *Bacillus*, nd-1.0×10^6; *Bacteroides* type A, 2.8×10^8–1.2×10^9; *Bacteroides* type B, 7.6×10^5–3.1×10^7; other *Bacteroidaceae*, 3.8×10^6–1.5×10^8; *Clostridium*, 7.6×10^5–2.7×10^7; and total viable counts, 2.9×10^8–1.5×10^9. On the other hand, the intestinal microflora (CFU/g) of channel catfish was: *Aeromonas*, 8.6×10^5–5.1×10^6; *Enterobacteriaceae*, nd-7.8×10^2; *Pseudomonas*, nd-1.7×10^6; *Acinetobacter*, nd-7.8×10^2; *Bacillus*, nd-1.8×10^5; *Bacteroidaceae*, nd-8.0×10^2; *Clostridium*, nd-2.0×10^3; and total viable counts, 2.2×10^5–6.8×10^8. This result, along with the previous report,[4] indicates that obligate anaerobes, especially *Bacteroides* type A, occur predominantly in the tilapia's intestine but poorly in the channel catfish's intestine.

Fig. 1 shows the vitamin B_{12}-producing ability (ng/ml/optical density at 630 nm) of bacterial strains. Of

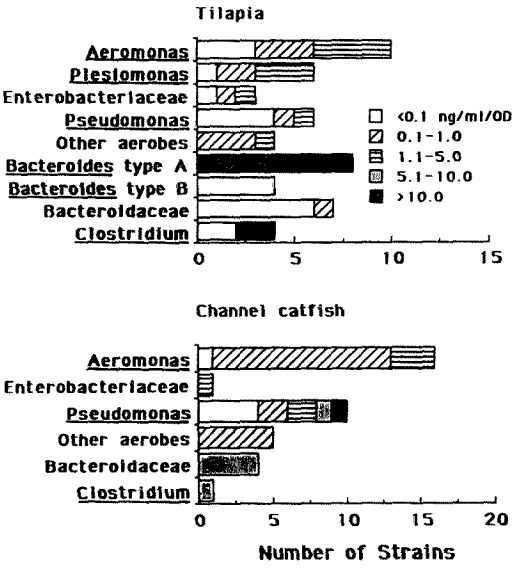

Fig. 1. The vitamin B_{12}-producing ability of intestinal bacteria from tilapia and channel catfish.

89 strains examined, 65 strains produced the vitamin B_{12} whilst the other 24 strains consumed it. Especially 17 strains, consisting of 100% of *Bacteroides* type A, 60% of *Clostridium*, 36% of other *Bacteroidaceae* and 20% of *Pseudomonas*, synthesized the vitamin B_{12} efficiently (>5.0 ng/ml/OD_{630}). These results strongly suggest that the obligate anaerobes, including *Bacteroides* type A, other *Bacteroidaceae* and *Clostridium*, are the imporant producer of this substance in the intestinal tract of freshwater fish. Therefore, the difference of the vitamin B_{12}-producing rate in the intestinal tract of tilapia and channel catfish may be attributed to the fact that the former harbours anaerobes abundantly whereas the later poorly. Additionally, the great difference of total viable counts between two fish species may be involved. Further studies along these lines are in progress.

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