Competition for attachment of aquaculture candidate probiotic and pathogenic bacteria on fish intestinal mucus

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Abstract

Probiotics for aquaculture are generally only selected by their ability to produce antimicrobial metabolites, however, attachment to intestinal mucus is important in order to remain within the gut of its host. Five candidate probiotics (AP1–AP5), isolated from the clownfish, *Amphiprion percula* (Lacepède), were examined for their ability to attach to fish intestinal mucus and compete with two pathogens, *Aeromonas hydrophila* and *Vibrio alginolyticus*. Two different radioactive isotopes were used to quantify competition between pathogens and probionts. Attachment of the pathogens was enhanced by the presence of the candidate probiotics. However, the addition of the candidate probiotics after the pathogens resulted in reduced pathogen attachment. Only AP5 caused lower attachment numbers of *V. alginolyticus* when added before the pathogen. When AP5 was added first, the average attachment change was 41% compared with 72% when added after *V. alginolyticus*, suggesting that the probiotic is displaced but that enhanced attachment of the pathogen does not occur. Conversely, when *V. alginolyticus* was added first, followed by AP5, attachment change was 37% while AP5 had 92% attachment change when added second. This implies that the pathogen was displaced by the candidate probiotic and therefore it appeared that, based on the ability of probiont AP5 to attach to mucus, the growth of the pathogen in the digestive tract might be suppressed by the candidate probiont's presence.

Keywords: *Aeromonas*, attachment, competition, intestinal mucus, probiotics, *Vibrio*.

Introduction

The worldwide increase in bacterial resistance to antibiotics (van der Waaij & Nord 2000) has stimulated investigations into the use of probiotics. In aquaculture, antibiotics are discharged into the environment causing the occurrence of resistant bacteria on fish farms (Aoki, Jo & Egusa 1980; DePaola 1995; Kerry, NicGabhainn & Smith 1997; Miranda & Zemelman 2002) or in the sediment below net cages (Bjorklund, Bondestam & Bylund 1990). With tighter regulations on the use of antibiotics in aquaculture, the use of probiotics has increasing potential (Austin, Stuckey, Robertson, Effendi & Griffith 1995; Ruiz, Roman & Sanchez 1996; Barker 1998; Gatesoupe & Lesel 1998; Gibson, Woodworth & George 1998; Rengpipat, Phianphak, Piyatiratitivorakul, Menasveta, Srirat, Wannipa, Somkiat & Pramote 1998; Shiri, Van Duffel, Dhert, Swings & Sorgeloos 1998; Gatesoupe 1999; Gram, Melchiorsen, Spanggaard, Huber & Nielsen 1999; Ringø & Birkbeck 1999; Skjermo & Vadstein 1999; Gomez-Gil, Roque & Turnbull 2000). Probiotics make up part of the resident microflora and contribute to the health or well-being of their host (Gatesoupe 1999). To remain within their host, they must either attach to the intestinal tract or grow fast enough to prevent them from being flushed out by the movement of

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