environmental microbiology

Environmental Microbiology (2013) **15**(7), 1917–1942 doi:10.1111/1462-2920.12134

Minireview

Antimicrobial use in aquaculture re-examined: its relevance to antimicrobial resistance and to animal and human health

Felipe C. Cabello,[1]* Henry P. Godfrey,[2] Alexandra Tomova,[1] Larisa Ivanova,[1] Humberto Dölz,[3] Ana Millanao[3] and Alejandro H. Buschmann[4]

Departments of [1]Microbiology and Immunology and [2]Pathology, New York Medical College, Valhalla, NY 10595, USA.

[3]Instituto de Farmacia, Facultad de Ciencias, Universidad Austral de Chile, Valdivia, Chile.

[4]Centro i~mar, Universidad de Los Lagos, Puerto Montt, Chile.

Summary

The worldwide growth of aquaculture has been accompanied by a rapid increase in therapeutic and prophylactic usage of antimicrobials including those important in human therapeutics. Approximately 80% of antimicrobials used in aquaculture enter the environment with their activity intact where they select for bacteria whose resistance arises from mutations or more importantly, from mobile genetic elements containing multiple resistance determinants transmissible to other bacteria. Such selection alters biodiversity in aquatic environments and the normal flora of fish and shellfish. The commonality of the mobilome (the total of all mobile genetic elements in a genome) between aquatic and terrestrial bacteria together with the presence of residual antimicrobials, biofilms, and high concentrations of bacteriophages where the aquatic environment may also be contaminated with pathogens of human and animal origin can stimulate exchange of genetic information between aquatic and terrestrial bacteria. Several recently found genetic elements and resistance determinants for quinolones, tetracyclines, and β-lactamases are shared between aquatic bacteria, fish pathogens, and human pathogens, and appear to have originated in aquatic bacteria. Excessive use of antimicrobials in aquaculture can thus potentially negatively impact animal and human health as well as the aquatic environment and should be better assessed and regulated.

Received 30 November, 2012; revised 10 February, 2013; accepted 14 February, 2013. *For correspondence. E-mail fcabello@nymc.edu; Tel. (914) 594 4182; Fax (914) 594 4176.

Introduction

Even though much of the rapid growth of aquaculture over the past quarter century has taken place in Asia (Arthur *et al*., 2000; Costa-Pierce, 2003; 2010; Naylor and Burke, 2005; Asche *et al*., 2008; Cole *et al*., 2009; Diana, 2009), development and application of intensive methods of salmon farming in Norway and Chile have resulted in their being among the top 12 aquacultural producers of animal protein in the world (Chopin *et al*., 2008; FAO, 2010). This widespread growth of aquaculture has been accompanied by an increased use of a wide range of chemicals including antimicrobials (Haya *et al*., 2001; Armstrong *et al*., 2005; Cabello, 2006; Buschmann *et al*., 2009; Cole *et al*., 2009; Asche *et al*., 2010; Burridge *et al*., 2010; Millanao *et al*., 2011). Increases in aquacultural antimicrobial use have been difficult to assess because of the large size and geographical extent of the industry, the various modalities employed (i.e. extensive, integrated, and intensive), and the over 200 species of fish and shellfish involved (Austin, 1985; Arthur *et al*., 2000; Costa-Pierce, 2003; 2010; Naylor and Burke, 2005; Asche *et al*., 2008; 2010; Asche, 2009; Diana, 2009). Collection of information about antimicrobial use in aquaculture is further complicated by a wide range of proprietorship (family units, village ownership, small businesses, international conglomerates) (Austin, 1985; Costa-Pierce, 2003; 2010; Naylor and Burke, 2005; Asche *et al*., 2008; Asche, 2009; Diana, 2009; Rodgers and Furones, 2009) as well as by differing national regulations which often do not encourage data collection for purposes of animal and public health and epidemiology (Asche *et al*., 2008; Asche, 2009; Burridge *et al*., 2010; Millanao *et al*., 2011).

Despite these impediments, available information has revealed widespread geographical heterogeneity in the amounts and classes of antimicrobials used in aquaculture (Burridge *et al*., 2010; Millanao *et al*., 2011; Ndi and Barton, 2012). It has also indicated that intensive aquaculture in some countries is an especially important source for passage of antimicrobials into the aquatic environment with potential effects on the health of fish, terrestrial animals, human beings, and the environment in general (Burridge *et al*., 2010; Millanao *et al*., 2011; Miranda, 2012). One of us has previously succinctly reviewed antimicrobial use in aquaculture and the implications of this use for biodiversity and human health (Cabello, 2006). This present more comprehensive review examines recently emerging and past information about antimicrobial use in aquaculture and its impact on the molecular genetics and evolution of antimicrobial resistance in the environment. Some aspects of this review concentrate on salmon aquaculture because of the availability of relatively reliable information obtained by us about this industry's usage of antimicrobials (Millanao, 2002; Barrientos, 2006; Gómez, 2009; Millanao *et al*., 2011) and because of the important potential impacts of this rapidly growing industry on aquatic biodiversity, antimicrobial resistance evolution, and piscine, terrestrial animal and human health.

Antimicrobial use in aquaculture

Classes and amounts

A large proportion, perhaps half, of the world's industrial production of antimicrobials is consumed in terrestrial animal agriculture; their use as prophylactics and as growth promoters far outweighs their use as therapeutics (Mellon *et al*., 2001; Sarmah *et al*., 2006; Davies, 2009; Davies and Davies, 2010; Levy and Marshall, 2010; Bush *et al*., 2011; Marshall and Levy, 2011). Antimicrobials are used in aquaculture not to promote growth but rather to prevent and treat bacterial infections in fish and invertebrates. These arise as a consequence of lowered host defences associated with culture at high density with suboptimal hygiene in enclosures in close proximity (Austin, 1985; Barton and Iwama, 1991; Grave *et al*., 1999; Arthur *et al*., 2000; Woo *et al*., 2002; Beveridge, 2004; Armstrong *et al*., 2005; Defoirdt *et al*., 2007; Sapkota *et al*., 2008; Grave and Hansen, 2009; Rodgers and Furones, 2009; Burridge *et al*., 2010; Millanao *et al*., 2011; Austin and Austin, 2012). These conditions, often associated with efforts to increase productivity, in turn favour development and epizootic dissemination of bacterial infections among aquaculture units in a geographical area (Barton and Iwama, 1991; Burka *et al*., 1997; Grave *et al*., 1999; Sørum, 2000; 2006; Woo *et al*., 2002; Beveridge, 2004; Cabello, 2006; Cole *et al*., 2009; Grave and Hansen, 2009; Asche *et al*., 2010; Barton and Floysand, 2010; Ibieta *et al*., 2011; Millanao *et al*., 2011). In salmon aquaculture, the need to grow different developmental stages in fresh and salt water and the manipulations to transport them between these two environments also increases stress and the opportunities for contact between different populations of fish, thus increasing opportunities for cross infection (Woo *et al*., 2002; Beveridge, 2004; Ibieta *et al*., 2011).

Aquacultural use of antimicrobials in developed countries has generally been restricted to avoid potential selection for human pathogens resistant to antimicrobials effective in clinical practice (Grave *et al*., 1999; Collignon *et al*., 2009; Grave and Hansen, 2009; Heuer *et al*., 2009; Burridge *et al*., 2010). Canada, Norway and the United States permit aquacultural use of oxytetracycline, Canada and Norway permit use of florfenicol, and Norway permits aquacultural use of quinolones (Table 1) (Grave *et al*., 1999; Sapkota *et al*., 2008; Rodgers and Furones, 2009; Burridge *et al*., 2010). Information regarding classes of antimicrobials used in aquaculture is undoubtedly incomplete even in industrialized countries because regulatory agencies have failed to collect this information (Sapkota *et al*., 2008; Burridge *et al*., 2010; Marshall and Levy, 2011).

The situation is more problematic in countries where control is less stringent or lacking (Sapkota *et al*., 2008; Burridge *et al*., 2010; Marshall and Levy, 2011; Millanao *et al*., 2011). In contrast to the United States, Norway and Canada, Chile, the second largest producer of cultured salmon after Norway, not only permits aquacultural use of oxytetracycline, florfenicol, and quinolones, but also

Table 1. Antimicrobials currently authorized for use in salmon aquaculture in various countries.[a]

	Oxytetracycline	Florfenicol	Sulfa/trimethoprim derivatives	Quinolones	Others
Canada	+	+	+		
Chile	+	+		+ (Oxolinic acid, Flumequin, others)	Amoxicillin, Erythromycin, Furazolidin Chloramphenicol, Gentamycin
Norway	+	+		+ (Oxolinic acid, Flumequin)	
United States	+		+		

a. Burridge *et al.* (2010)

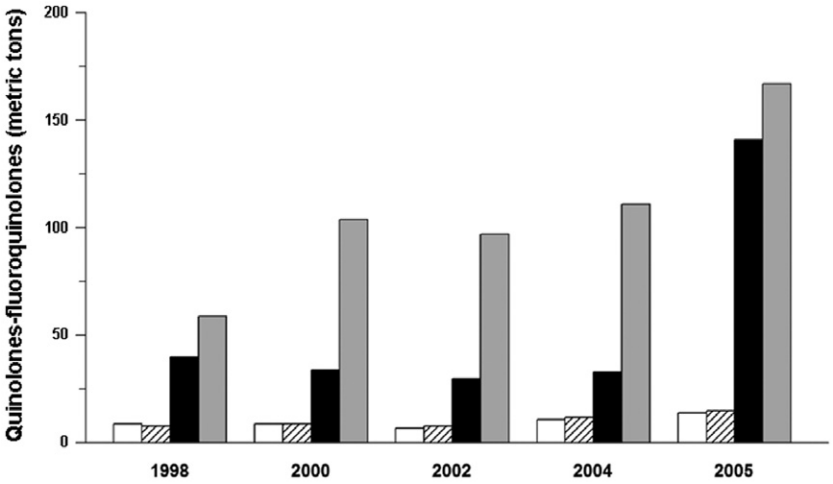

Fig. 1. Authorized and imported quantities (metric tons) of quinolones and fluoroquinolones for human and veterinary medical use in Chile, 1998–2005. Quinolones-fluoroquinolones authorized for import for human use by the Chilean Institute of Public Health (white bars) and actually imported (hatched bars). Quinolones-fluoroquinolones authorized for import for veterinary use by the Chilean Agricultural and Livestock Service (black bars) and actually imported (grey bars). Modified from (Millanao, 2002; Barrientos, 2006; Millanao *et al*., 2011; A. Millanao and H. Dölz, unpublished).

allows use of amoxicillin, erythromycin and several other antimicrobials (Table 1) (Grave *et al*., 1999; Sapkota *et al*., 2008; Grave and Hansen, 2009; Rodgers and Furones, 2009; Burridge *et al*., 2010). According our own investigations, agricultural regulators in Chile have consistently failed to successfully track and limit veterinary use of antimicrobials (Millanao, 2002; Barrientos, 2006; Millanao *et al*., 2011). Between 1998 and 2004, 1.5 to 3.4 times more quinolones and fluoroquinolones were imported into Chile for veterinary medicine and used preferentially in aquaculture than were authorized by the national Livestock and Agricultural Service (Fig. 1). This ratio fell to 1.2 in 2005 (although use was still high) perhaps because permits were increased (Fig. 1), suggesting there was previously an unregulated market for veterinary use of these drugs (Millanao, 2002; Barrientos, 2006; Millanao *et al*., 2011; A. Millanao and H. Dölz, unpublished). Quinolones such as oxolinic acid and flumequine comprise most of the quinolones imported to Chile and together with florfenicol are mostly used in aquaculture (Millanao, 2002; Barrientos, 2006; Millanao *et al*., 2011). Studies of antimicrobial resistance in lower intensity fish and shrimp aquacultural settings also suggest that many classes of antimicrobials are employed in these activities as well (Holmström *et al*., 2003; Le and Munekage, 2004; Le *et al*., 2005; Hastein *et al*., 2006). Detection of nitrofurans by the Food and Drug Administration in aquacultural products imported to the United States from China (Burridge *et al*., 2010; Love *et al*., 2011), and detection of chloramphenicol and metronidazole by the European Union regulatory authorities in seafood imported from China, Indonesia, Taiwan, Thailand and Vietnam provide additional evidence for lax control of antimicrobial use in other less industrialized countries (Rodgers and Furones, 2009; Love *et al*., 2011).

There is a great variability in the amounts and classes of antimicrobials used in salmon, shrimp and other forms of aquaculture from country to country (Grave *et al*., 1999; 2006; Holmström *et al*., 2003; Le and Munekage, 2004; Grave and Hansen, 2009; Burridge *et al*., 2010; Millanao *et al*., 2011). For example, Japanese aquaculture used 179 metric tons of antimicrobials in 2001, slightly more than a third of the amount used in human medicine that year (Furushita and Shiba, 2007). The situation was quite different in intensive salmon aquaculture in Chile. Importation of antimicrobials to Chile in the period 2000–2007 for use in veterinary medicine increased in parallel with salmonid production (Figs 1 and 2). It was several times greater than importation of antimicrobials for human medicine which increased only slightly over this time (Millanao, 2002; Barrientos, 2006; Gómez, 2009; Millanao *et al*., 2011). While Norway, the United Kingdom and Canada used approximately 0.0008 kg, 0.0117 kg, and 0.175 kg, respectively, of antimicrobials for each metric ton of salmon produced in 2007, Chile used at least 1.4 kg per metric ton (Fig. 2) (SalmonChile, 2008; Gómez, 2009; Burridge *et al*., 2010; Millanao *et al*., 2011; A. Millanao and H. Dölz, unpublished). Thus, considerably more antimicrobials were used in Chile than in Norway or Canada to produce one metric ton of salmon (approximately over 1500 and eight times more respectively) (Millanao, 2002; Barrientos, 2006; Gómez, 2009; Burridge *et al*., 2010; Millanao *et al*., 2011; A. Millanao and H. Dölz, unpublished). Approximately 471, 233, and 226 metric tons of tetracycline, florfenicol, and quinolones, respectively, were estimated to have been used in Chile in 2007 in veterinary medicine (Millanao, 2002; Barrientos, 2006; Millanao *et al*., 2011). These 930 metric tons of antimicrobials were mostly used in salmon aquaculture (Millanao, 2002; Barrientos, 2006; Gómez, 2009; Millanao *et al*., 2011).

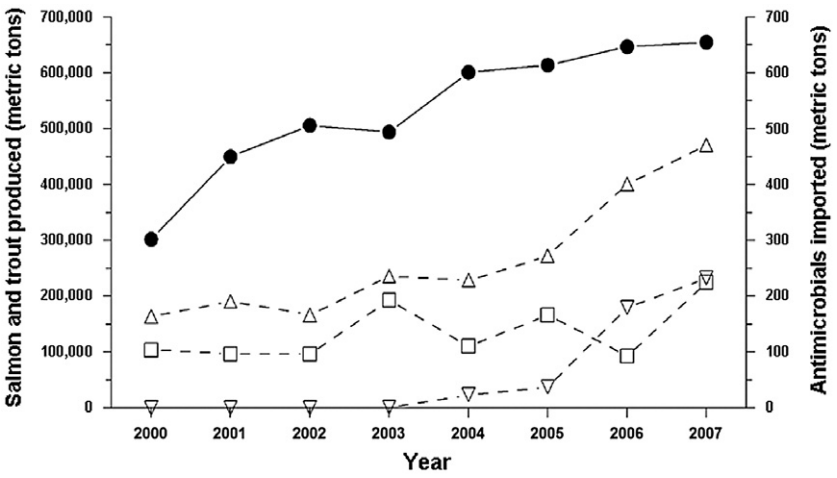

Fig. 2. Production of salmon and trout in Chile and net imports of selected antimicrobials for veterinary use (both in metric tons) to Chile, 2000–2007. Production of salmon and trout (●); imports of tetracyclines (Δ), quinolones/fluoroquinolones (□), and florfenicol (∇). Modified from (Millanao, 2002; Barrientos, 2006; SalmonChile, 2008; Gómez, 2009; Millanao *et al*., 2011; A. Millanao and H. Dölz, unpublished).

Antimicrobials in water and sediments

Antimicrobials used in aquaculture are administered to fish mostly in food and only rarely by injection or bath (Capone *et al*., 1996; Herwig *et al*., 1997; Armstrong *et al*., 2005; Sørum, 2006; Rodgers and Furones, 2009). This method of administration leads to their affecting both diseased and healthy fish (metaphylaxis) in the population (Sørum, 2006). Unconsumed medicated food (perhaps as much as 30% of that supplied if fish are diseased and anorexic) is deposited by gravity in sediments under and around aquaculture sites (Björklund *et al*., 1990; Capone *et al*., 1996; Herwig *et al*., 1997; Armstrong *et al*., 2005; Sarmah *et al*., 2006; Sørum, 2006; Sapkota *et al*., 2008; Pelletier *et al*., 2009; Rodgers and Furones, 2009). Of the ingested antimicrobials, approximately 80% pass into the environment in unabsorbed form in faeces or after absorption, in secreted forms in urine and other secretions (Björklund *et al*., 1990; Hektoen *et al*., 1995; Capone *et al*., 1996; Burka *et al*., 1997; Le and Munekage, 2004; Armstrong *et al*., 2005; Sørum, 2006). These also accumulate in the sediments under and around the aquaculture pens (Björklund *et al*., 1990; Hektoen *et al*., 1995; Capone *et al*., 1996; Arthur *et al*., 2000; Coyne *et al*., 2001; Le and Munekage, 2004; Armstrong *et al*., 2005) from where they can be carried by water currents to sediments at distant sites (Samuelsen *et al*., 1992b; Capone *et al*., 1996; Coyne *et al*., 1997; 2001; Arthur *et al*., 2000; Fortt *et al*., 2007; Buschmann *et al*., 2012). In places where hundreds of metric tons of antimicrobials are used per year in a limited geographical area, antimicrobials may remain in large amounts for far longer periods of time than was previously thought to occur (Asche *et al*., 2010; Burridge *et al*., 2010; Millanao *et al*., 2011; Buschmann *et al*., 2012). Antimicrobials leached from sediments as well as from ingestion of uneaten medicated feed can also potentially affect free-ranging fish, shellfish and other animals in proximity to aquaculture sites (Björklund *et al*., 1990; Samuelsen *et al*., 1992b; Capone *et al*., 1996; Coyne *et al*., 1997; Fortt *et al*., 2007).

The length of time untransformed and transformed antimicrobial activity remains in sediments is dependent on the initial concentrations of antimicrobials (i.e. proportional to the total amounts used at aquaculture sites), their chemical structures, and the half-life of these compounds (Björklund *et al*., 1990; 1991; Husevåg *et al*., 1991; Samuelsen *et al*., 1994; Hektoen *et al*., 1995; Capone *et al*., 1996; Kerry *et al*., 1996; Arthur *et al*., 2000; Chelossi *et al*., 2003; Boxall *et al*., 2004; Kummerer, 2009). Environmental chemical and physical variables such as sediment characteristics, water currents, temperature, light and pH also influence the length of time sediments retain antimicrobial activity (Capone *et al*., 1996; Kummerer, 2009). Leaching into water and dispersion by currents appears to be the main mechanism mediating decreases in antimicrobial concentrations in sediments rather than degradation *per se*, but this has not been extensively studied (Björklund *et al*., 1990; Samuelsen *et al*., 1992a; 1994; Hektoen *et al*., 1995; Kummerer, 2009). Field and laboratory investigations have indicated that detectable concentrations of biologically-active oxytetracycline remain in sediments for months to more than a year (Björklund *et al*., 1990; Hektoen *et al*., 1995; Capone *et al*., 1996; Coyne *et al*., 2001; Koeypudsa *et al*., 2005). Studies on artificial marine sediments suggest that non-degradable quinolones such as oxolinic acid and flumequine may persist close to aquaculture sites months after their utilization (Hansen *et al*., 1993; Samuelsen *et al*., 1994; Hektoen *et al*., 1995; Lai and Lin, 2009). Similar studies with sulfa drugs, trimethoprim and florfenicol also suggest that these remain active in sediments for several months (Samuelsen *et al*., 1994; Hektoen *et al*., 1995; Capone *et al*., 1996; Hoa

et al., 2008). Although florfenicol disappears in a few days, one of its derivatives, florfenicol amine, remains in sediments for months (Hektoen *et al*., 1995). The half-life of antimicrobials within and under sediments is also prolonged; they continue to be able to exert selective pressure in this location for an extended period of time (Hektoen *et al*., 1995; Capone *et al*., 1996). Antimicrobials such as tetracycline can exert antimicrobial activity even if they adsorb to sediments and react with inhibitory cations such as Mg^{2+} and Ca^{2+}, especially in areas where large amounts are used and subinhibitory concentrations are maintained in the environment (Barnes *et al*., 1995; Lunestad and Goksøyr, 2010). Some authors have claimed that antimicrobials such as tetracycline do not end up in sediments because only minimal amounts are detectable there (Smith, 1996; Miranda, 2012). The subinhibitory concentrations of antimicrobials in the sediment postulated by supporters of this hypothesis would still have sufficient biological activity to affect horizontal gene transfer (HGT) and mutagenesis in bacteria (Beaber *et al*., 2004; Hastings *et al*., 2004; Davies, 2009; Gullberg *et al*., 2011). In fact, concentrations of antimicrobials detected in sediments in several studies are still many times greater than the minimal inhibitory concentrations for most bacteria (Samuelsen, 1989; Björklund *et al*., 1991; Samuelsen *et al*., 1992a; Capone *et al*., 1996; Smith, 1996; Tello *et al*., 2012).

Effects of antimicrobials in the aquacultural environment

Selection of antimicrobial-resistant bacteria

Significant concentrations of antimicrobials remaining for long periods of time in the aquatic environment are the principal selective pressure for antimicrobial resistance in bacteria in sediments and the overlying water column (Samuelsen *et al*., 1994; Hektoen *et al*., 1995; Capone *et al*., 1996; Herwig *et al*., 1997; Petersen *et al*., 2002; Giraud *et al*., 2006; Dang *et al*., 2007; Baquero *et al*., 2008; 2009; Ding and He, 2010; Marshall and Levy, 2011). The impact of this process leads to a major alteration of the biodiversity of the sediment and water by replacing susceptible communities of bacteria and other microorganisms with resistant ones. This impact has been extensively documented both in the laboratory and in the field (DePaola *et al*., 1995; Capone *et al*., 1996; Herwig and Gray, 1997; Herwig *et al*., 1997; Holten Lützhøft *et al*., 1999; Arthur *et al*., 2000; Guardabassi *et al*., 2000; Miranda and Zemelman, 2002a,b; Kim *et al*., 2004; 2011; Le and Munekage, 2004; Alcaide *et al*., 2005; Le *et al*., 2005; Akinbowale *et al*., 2006; 2007; Christensen *et al*., 2006; Giraud *et al*., 2006; Cordova-Kreylos and Scow, 2007; Dang *et al*., 2007; 2011; Gonçalves Ferreira *et al*., 2007; Gordon *et al*., 2007; Miranda and Rojas, 2007; Heepngoen *et al*., 2008; American Academy of Microbiology, 2009; Ding and He, 2010; Fernández-Alarcón *et al*., 2010; Ishida *et al*., 2010; Andersson and Hughes, 2011). Significant increases in the frequency of bacteria resistant to oxytetracycline, quinolones, sulfa/trimethoprim, florfenicol, and amoxicillin have been repeatedly found in proximity to aquaculture farms employing these antimicrobials, suggesting a causal relationship between these variables (DePaola *et al*., 1995; Guardabassi *et al*., 2000; Schmidt *et al*., 2000; Dang *et al*., 2007; Gordon *et al*., 2007; Suzuki, 2010). Moreover, antimicrobial-resistant bacteria are found at aquaculture sites for a prolonged period of time after antimicrobial use, further suggesting the relevance of this selection over time (Husevåg *et al*., 1991; Tamminen *et al*., 2011b). Laboratory models using aquatic sediments have consistently demonstrated that introduction of antimicrobials is accompanied by increases in the frequency of antimicrobial-resistant bacteria (Hansen *et al*., 1993; Herwig and Gray, 1997; Stepanauskas *et al*., 2006) and, as expected from the modular clustering of antimicrobial resistant genetic elements, introduction of one antimicrobial can give rise to bacteria resistant to other antimicrobials that are not even in use in the area (Herwig and Gray, 1997; Le *et al*., 2005; Alekshun and Levy, 2007; Stokes and Gillings, 2011). Whether these antimicrobials remain in the sediment or leach into the surrounding water, the end result is still selection of antimicrobial-resistant bacteria (Davies and Davies, 2010; Marshall and Levy, 2011; Buschmann *et al*., 2012).

The fact that Chilean salmon aquaculture experienced epizootics and infestations resulting from unsanitary conditions strongly suggests that a large proportion of these antimicrobials were used for prophylaxis rather than for therapeutics (Godoy *et al*., 2008; Kibenge *et al*., 2009; Asche *et al*., 2010; Ibieta *et al*., 2011; Millanao *et al*., 2011). In Chile at least, aquaculture rather than human and other veterinary medical activities would seem to be the most important source for passage of antimicrobials into the aquatic environment where they select for antimicrobial-resistant bacteria (Asche *et al*., 2010; Ibieta *et al*., 2011; Millanao *et al*., 2011). In view of the continuing worldwide increase in aquaculture, the effects of antimicrobial use in this industry raise questions that deserve careful monitoring (FAO, 2010).

The emergence of antimicrobial-resistant bacteria may even be greater than that which has been detected since most studies have been limited to demonstrating this resistance in culturable bacteria, which constitute only a small proportion of the total bacteria present in the aquatic environment (Bissett *et al*., 2006). There is a lack of information regarding microbial communities that change in numbers or even disappear in aquatic environments

because of their susceptibility to antimicrobials and the effect this phenomenon may have on metabolic activities of microbial communities and the health of the sediment (Bissett *et al*., 2006; Edlund *et al*., 2006; Ma *et al*., 2006; Gonçalves Ferreira *et al*., 2007). Deposition of food pellets and organic matter lacking antimicrobials onto sediments near aquaculture sites and in the laboratory have been shown to impact sediment microbial biodiversity and have been suggested to increase the fraction of antimicrobial-resistant bacteria present in them (Smith *et al*., 1994; Kapetanaki *et al*., 1995; Nogales *et al*., 2011; Pitkanen *et al*., 2011; Tamminen *et al*., 2011a). However, these studies did not rule out the presence of other antimicrobial compounds in the food or in the sediments such as heavy metals (Zn, Cu, Hg), disinfectants, and organic antibacterial compounds, any or all of which might be responsible for these results (Smith *et al*., 1994; Kapetanaki *et al*., 1995; Akinbowale *et al*., 2007; Tacon and Metian, 2008; Pitkanen *et al*., 2011; Tamminen *et al*., 2011a). Although the observed increase in antimicrobial-resistant bacteria could be explained by linkage of genes involved in metabolism of organic matter with antimicrobial resistance genes, the preponderance of evidence to date suggests that antimicrobial residues present in the environment where aquaculture takes place are the most relevant selective pressure to account for the increased fraction of antimicrobial-resistant bacteria there.

Mechanisms of bacterial selection

The genomes of aquatic bacteria are highly diverse and contain genetic elements and genes involved in the generation and dissemination of antimicrobial resistance genes similar to those previously characterized in terrestrial bacteria (Venter *et al*., 2004; Baker-Austin *et al*., 2009; Biers *et al*., 2009; Sobecky and Hazen, 2009; Hazen *et al*., 2010; McDaniel *et al*., 2010; Wiedenbeck and Cohan, 2011). The total of all mobile genetic elements (MGE) of the genome of aquatic bacteria, the mobilome, include water current-transported naked DNA (Stewart and Sinigalliano, 1990; Sobecky and Hazen, 2009; Fondi and Fani, 2010; Taylor *et al*., 2011; Domingues *et al*., 2012), insertion sequences (Toleman and Walsh, 2011), insertion sequence elements with common regions (IS*CR*) (Toleman *et al*., 2006; Toleman and Walsh, 2010; Xia *et al*., 2010), integrons mobilized by plasmids, transposons and integrative and conjugative elements (ICE or SXT) (Rosser and Young, 1999; L'Abee-Lund and Sørum, 2001; Schmidt *et al*., 2001b; Burrus *et al*., 2006; Koenig *et al*., 2008; Osorio *et al*., 2008; Wozniak *et al*., 2009; Cambray *et al*., 2010; Daccord *et al*., 2010; Rosewarne *et al*., 2010; Wozniak and Waldor, 2010), genomic islands (Boyd *et al*., 2002; 2008; Juhas *et al*., 2009; Daccord *et al*., 2010; Le Hello *et al*., 2011), transposons and conjugative transposons (Rhodes *et al*., 2000; Knapp *et al*., 2008), conjugative and mobilizable plasmids (Baya *et al*., 1986; Aoki *et al*., 1987; Kim and Aoki, 1996b; Sobecky *et al*., 1997; Schmidt *et al*., 2001b; Furushita *et al*., 2003; Kim *et al*., 2004; Rhodes *et al*., 2004; Gordon *et al*., 2007; Cattoir *et al*., 2008; Guglielmetti *et al*., 2009; Sobecky and Hazen, 2009; Erauso *et al*., 2011; Ma *et al*., 2012), and bacteriophages, including phage-like elements designated gene transfer agents (GTA) (Suttle, 2007; Colomer-Lluch *et al*., 2011; Lang *et al*., 2012). GTA mediate HGT between heterologous bacteria and appear to have an important role in this process in marine bacterial communities (Lang *et al*., 2012). It is not surprising that introduction of large amounts of antimicrobials into the aquatic environment is rapidly followed by emergence of significant numbers of multiple-resistant bacteria since antimicrobial resistance genes would enhance fitness for growth in sediments containing antimicrobials (Capone *et al*., 1996; Kerry *et al*., 1996; Sobecky *et al*., 1997; Guardabassi *et al*., 2000; Schmidt *et al*., 2000; Furushita *et al*., 2003; Groh *et al*., 2007; Seyfried *et al*., 2010). Moreover, contrary to well-documented reports showing that some antimicrobial resistance mechanisms have a fitness cost, the presence of the quinolone resistance gene *qnrA* in some aquatic bacteria and other antimicrobial resistance genes in *Shewanella* may enhance fitness in the absence of antimicrobials (Groh *et al*., 2007; Michon *et al*., 2011).

Conditions in aquatic environments that favour HGT include biofilms of aquatic bacteria on the epilithon (particulate organic matter coating benthic ecosystems), on clays and sand of sediments, and on aquacultural structures, coupled with the large concentrations of bacteriophages and GTAs in seawater, also favour HGT and dissemination of antimicrobial resistance (Hill *et al*., 1992; Sobecky *et al*., 1997; Bushman, 2002; Venter *et al*., 2004; Furushita and Shiba, 2007; Suttle, 2007; McDaniel *et al*., 2010; Marshall *et al*., 2011; Sundell and Wiklund, 2011; Taylor *et al*., 2011; Lang *et al*., 2012; Lupo *et al*., 2012; Toussaint and Chandler, 2012). Antimicrobials can potentially also stimulate HGT mediated by naked DNA generated by bacteriophage lysis, as well as that mediated by plasmids in the aquatic environment and in the intestines of fish and terrestrial animals (Stewart and Sinigalliano, 1990; Beaber *et al*., 2004; Frost *et al*., 2005; Aarestrup *et al*., 2008; Allen *et al*., 2011; Domingues *et al*., 2012; Looft *et al*., 2012). In addition, aquatic bacteriophages can contain antimicrobial resistance genes that may be expressed upon infection of bacteria (Colomer-Lluch *et al*., 2011). Several aquatic bacteria such as *Vibrio* spp. are naturally competent for DNA uptake, thus also increasing the opportunities for transformation to occur in the aquatic environment (Stewart and Sinigalliano, 1990; Meibom *et al*., 2005; Baharoglu *et al*., 2012).

Bacteria from aquatic and terrestrial environments share similar antimicrobial genetic determinants (Table 2, Fig. 3) (Baquero *et al*., 2008; Sobecky and Hazen, 2009; Marshall and Levy, 2011; Taylor *et al*., 2011; Buschmann *et al*., 2012), and HGT and recombination of these determinants between different bacterial species can be stimulated by residual and subinhibitory antimicrobial concentrations of tetracyclines and quinolones in sediments (Kruse and Sørum, 1994; Aarestrup *et al*., 2000; Beaber *et al*., 2004; Hastings *et al*., 2004; Davies, 2009; Buschmann *et al*., 2012). Bacteria in aquatic environments may in fact be the source of genetic elements of the mobilome such as SXT, IS*CR*, and integrons as well as previously unknown antimicrobial resistance determinants (Miranda *et al*., 2003; Burrus *et al*., 2006; Laroche *et al*., 2009; Daccord *et al*., 2010; Kristiansson *et al*., 2011; Xu *et al*., 2011a,b; Ma *et al*., 2012). For example, *tetG* (Table 2), an independently evolved tetracycline resistance determinant, was first discovered in aquatic bacteria (Aoki *et al*., 1987; Zhao and Aoki, 1992; Angulo, 1999). Several phenotypically tetracycline-resistant bacteria isolated from aquaculture sites also contained genetic determinants that could not be amplified by PCR with primers corresponding to the known tetracycline resistance determinants indicating that they carried unknown tetracycline resistance genes (Miranda *et al*., 2003).

A number of antimicrobial resistance genes appear to have been first detected in aquatic bacteria before being detected and disseminating among human and animal pathogens. These include some of the emerging plasmid-mediated quinolone resistance (PMQR) genes found in aquatic *Vibrio*, *Shewanella* and *Aeromonas* (Table 2) (Poirel *et al*., 2005; 2012; Cattoir *et al*., 2007; 2008; Xia *et al*., 2010); new β-lactamase genes from *Photobacterium damselae* (Table 2) (Morii, 2004) and *Oceanobacillus iheyensis* (Toth *et al*., 2010); a novel fosfomycin resistance determinant isolated from the aquatic environment (Xu *et al*., 2011b); the widely disseminated emerging *floR* gene of human pathogens (Kim and Aoki, 1996a; Angulo, 1999; Arcangioli *et al*., 1999; 2000; Bolton *et al*., 1999; Cloeckaert *et al*., 2000; 2001; Miranda and Rojas, 2007; Gordon *et al*., 2008; Smith, 2008a,b; Cabello, 2009; Welch *et al*., 2009; Fernández-Alarcón *et al*., 2010; Hall, 2010); and the chloramphenicol resistance genes *catII*, *catB9* and *catB2* from aquatic *Photobacterium, Vibrio* and *Shewanella* respectively (Roberts and Schwarz, 2009; Roberts *et al*., 2012). Moreover, antimicrobial resistance gene variants including those for β-lactams, aminoglycosides, tetracyclines, macrolides and heavy metals have been detected in the genome of the salmon pathogen *Renibacteriun salmoninarum* and the aquatic opportunistic human pathogen *Stenotrophomonas maltophilia* suggesting that these aquatic bacteria may be repositories for antimicrobial resistance genes (Crossman *et al*., 2008; Wiens *et al*., 2008).

Selection of antimicrobial-resistant bacteria in the aquatic environment can also occur by selection of spontaneous single mutants since water, sediments and piscine intestines all contain sufficiently large concentrations of bacteria to have detectable numbers of spontaneously arising antimicrobial-resistant mutants (Capone *et al*., 1996; Levy and Marshall, 2004; Alekshun and Levy, 2007; Navarrete *et al*., 2008; Navarro *et al*., 2008; Nikaido, 2009). Moreover, the high density of fish and shellfish in aquacultural enclosures increases the opportunities for this selection to occur (Woo *et al*., 2002; Beveridge, 2004; Austin and Austin, 2012). Mutants and bacteria tolerant to antimicrobials can clearly be selected by inhibitory and subinhibitory concentrations of antimicrobials (Miller *et al*., 2004; Dorr *et al*., 2009; Kohanski *et al*., 2010). Though this mechanism may not be as effective for selection and dissemination of antimicrobial-resistant bacteria as selection of bacteria containing MGEs with multiple antimicrobial resistance genes (Akinbowale *et al*., 2007; Davies, 2009; Davies and Davies, 2010), it may be relevant since persistent residual and subinhibitory concentrations of antimicrobials in sediments can trigger the SOS system (a bacterial reparative response to DNA damage). This system can increase the rate of mutagenesis by several mechanisms including generation of oxygen radicals (Kohanski *et al*., 2007; 2010; Dorr *et al*., 2009; Blazquez *et al*., 2012). Subinhibitory concentrations of antimicrobials can also select resistant bacteria by non-SOS-mediated mechanisms such as DNA recombination, amplification, and selection for hypermutator strains (López *et al*., 2007; Sun *et al*., 2009; Blazquez *et al*., 2012). These mechanisms may be especially relevant with quinolones located in sediments since these antibacterial agents are only slowly degraded and are well-known inducers of mutagenesis and antimicrobial tolerance (Dorr *et al*., 2009; Lai and Lin, 2009; Kohanski *et al*., 2010; Blazquez *et al*., 2012).

Antimicrobial-resistant mutants selected in fish intestinal tracts and in the environment can also have their mutated genes captured by integrons, genetic elements with diverse antimicrobial resistance determinant cassettes that can be mobilized by transposons and plasmids to generate new permutations of resistance genes (Rowe-Magnus and Mazel, 1999; L'Abee-Lund and Sørum, 2001; Mazel, 2006; Boucher *et al*., 2007; Jacobs and Chenia, 2007; Gillings *et al*., 2008; Laroche *et al*., 2009; Xia *et al*., 2010; Stalder *et al*., 2012). The frequent presence of integrons in aquatic bacteria, especially in bacteria from sediments impacted by anthropogenic activities such as aquaculture, may suggest an aquatic origin (Rosser and Young, 1999; Schmidt

Table 2. Some genetic elements and antimicrobial resistance genes shared between aquatic bacteria and human pathogens.

	β-proteobacteria	*Shewanella*	*Vibrio*	*Photobacterium*	*Aeromonas*	*Moraxella*	*Acinetobacter*	*Y. ruckeri*	*Kluyvera*	Reference
Elements										
ICE		+	+	+						a
Integron 1	+	+	+							b
IS*CR*		+	+	+	+					c
SXT/391		+	+	+						d
pIncA/C			+	+	+			+		e
pIncU				+	+					f
SGI1				+						g
Tn*1721*					+					h
Genes										
floR				+	+					i
β-lactamase				+						j
CTX-*β-lactamase*									+	k
qnrA		+								l
qnrS			+	+	+					m
qnrVC			+		+					n
tetD (E)			+		+					o
tetH						+	+			p
tetC					+					q
tetG			+							r

a. Burrus *et al*. (2006); Osorio *et al*. (2008).; Toleman and Walsh (2011); Rodriguez-Blanco *et al*. (2012).
b. L'Abee-Lund and Sørum (2001); Boucher *et al*. (2007); Gillings *et al*. (2008); Koenig *et al*. (2008); Laroche *et al*. (2009); Cambray *et al*. (2010).
c. Toleman *et al*. (2006); Toleman and Walsh (2011).
d. Wozniak *et al*. (2009); Wozniak and Waldor (2010).
e. McIntosh *et al*. (2008); Fricke *et al*. (2009); Welch *et al*. (2009).
f. Sørum *et al*. (2003); Rhodes *et al*. (2004).
g. Briggs and Fratamico (1999); Boyd *et al*. (2008).
h. Rhodes *et al*. (2000).
i. Kim and Aoki (1996a); Arcangioli *et al*. (1999); Cloeckaert *et al*. (2000); Gordon *et al*. (2008); Smith (2008a,b); Cabello (2009); Welch *et al*. (2009).
j. Morii (2004).
k. Decousser *et al*. (2001); Rodriguez *et al*. (2004); Cantón and Coque (2006); Cantón *et al*. (2012).
l. Poirel *et al*. (2005).
m. Saga *et al*. (2005); Cattoir *et al*. (2007); Cattoir *et al*. (2008); Poirel *et al*. (2012).
n. Xia *et al*. (2010).
o. Furushita *et al*. (2003); Roberts and Schwarz (2009).
p. Miranda *et al*. (2003).
q. Furushita *et al*. (2003); Sandoz and Rockey (2010).
r. Zhao and Aoki (1992).

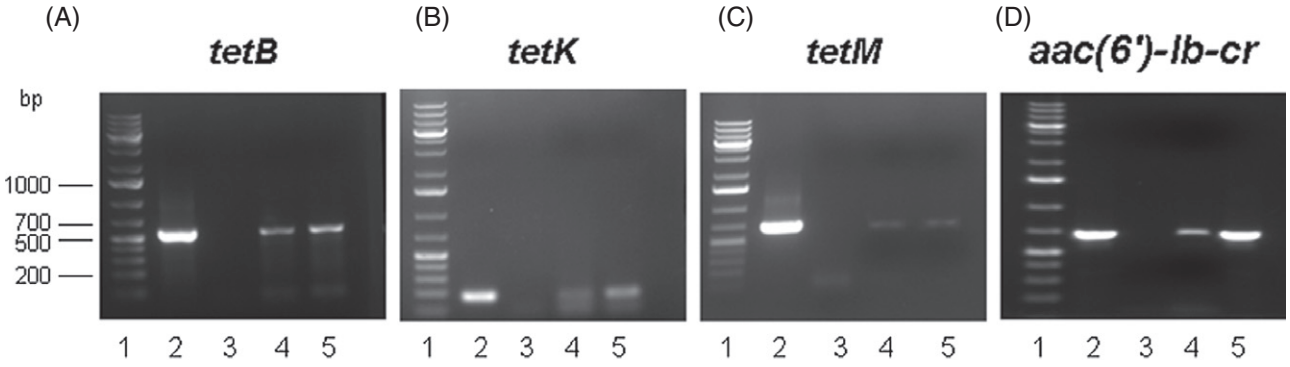

Fig. 3. Sharing of tetracycline and plasmid mediated quinolone resistance determinant *aac(6′)-Ib-cr* between marine bacteria and four *Escherichia coli* clinical isolates (isolates 1–4) from human urinary tract infections in Region X, Chile. Amplicons of *tetB*, *tetK*, *tetM* and *aac(6′)-Ib-cr* were detected by PCR as described (Ng *et al*., 2001; Miranda *et al*., 2003; Robicsek *et al*., 2006). For all panels, lane 1, molecular weight standards; lane 2, positive control; lane 3, negative control (*E. coli* DH5α); lane 4, marine bacterial isolate; lane 5, clinical isolate. A. *tetB*. Lane 2, *E. coli* DH5α pET*tetB1*; lane 4, *Pseudoalteromonas* sp.; lane 5, *E. coli* isolate 1. B. *tetK*. Lane 2, *S. aureus* pT181; lane 4, *Pseudoalteromonas* sp.; lane 5, *E. coli* isolate 2. C. *tetM*. Lane 2, *E. coli* DH10B pJFP76; lane 4, *Shewanella* sp.; lane 5, *E. coli* isolate 3. D. *aac(6′)-Ib-cr*. Lane 2, *E. coli* J53 pMG298; lane 4, *Rhodoccus* sp.; lane 5, *E. coli* isolate 4.

et al., 2001a; Rosewarne *et al*., 2010; Gaze *et al*., 2011). Stimulation of the SOS stress regulon by antimicrobials such as quinolones and β-lactamases can not only stimulate HGT by transformation and conjugation but can also affect integron recombination and plasticity. This latter is a result of the triggering of integrase activity with its resultant antimicrobial resistance cassete uptake and expression (Baharoglu *et al*., 2010; 2012; Cambray *et al*., 2011). Subinhibitory concentrations of antimicrobials from aquacultural activities could thus, besides selecting and inducing antimicrobial resistance in sediments and water, also mediate antimicrobial resistance genetic plasticity *in vivo* in the intestine of aquaculture species (Guerin *et al*., 2009; Baharoglu *et al*., 2010; Baharoglu and Mazel, 2011; Blazquez *et al*., 2012; Hocquet *et al*., 2012).

It has been suggested that antimicrobial resistance genes and antimicrobial-resistant bacteria arrive there in fish food and in exogenous contaminating effluents rather than being generated from local sources in the water and sediments at aquaculture sites (Smith *et al*., 1994; Kapetanaki *et al*., 1995; Kerry *et al*., 1995; Smith, 2008b; Martinez, 2009b; 2012; Pitkanen *et al*., 2011). This and the fact that fish food may increase antimicrobial resistance (Smith *et al*., 1994; Kapetanaki *et al*., 1995; Kerry *et al*., 1995; Martinez, 2009b; Pitkanen *et al*., 2011) are plausible hypotheses that deserve investigation. However, persistence and increase of these genes and these bacteria in aquatic environments will be sustained by the presence of antimicrobials no matter how they arrive (Akinbowale *et al*., 2007; Davies, 2009; Nikaido, 2009; Davies and Davies, 2010). The modular nature of the MGE involved in antimicrobial resistance in aquatic bacteria also facilitates selection of multiple antimicrobial resistances by a single antimicrobial compound and by other antimicrobial compounds used in aquaculture such as heavy metals and disinfectants (Herwig and Gray, 1997; Lawrence, 2000; Stepanauskas *et al*., 2006; Akinbowale *et al*., 2007; Alekshun and Levy, 2007; Davies, 2009; Seiler and Berendonk, 2012).

Antimicrobial resistance genes have been demonstrated in ancient bacterial DNA extracted from terrestrial permafrost and in collections of bacteria preceding introduction of antimicrobials (Datta and Hughes, 1983; Hughes and Datta, 1983; D'Costa *et al*., 2011). While the effects of aquacultural antimicrobial use on aquatic sediments are most likely restricted to selecting those bacteria able to survive in their presence, the increase of antimicrobial-resistant bacteria that this produces and the particularities of the aquatic environment at aquaculture sites may provide new avenues for the generation and emergence of previously unknown and undescribed mechanisms for this selection as well as of new permutations of antimicrobial resistance genes as those generated by integron plasticity (Levesque *et al*., 1995; Sobecky *et al*., 1997; Sørum, 2006; Baquero *et al*., 2008; Sobecky and Hazen, 2009; Baharoglu *et al*., 2010; Cambray *et al*., 2010; Taylor *et al*., 2011; Hocquet *et al*., 2012). In aquaculture and the aquatic environment, antimicrobials clearly appear to display their hormetic properties: higher concentrations of antimicrobials select for resistant bacteria, while subinhibitory concentrations of their residues might stimulate HGT and mutagenesis (Linares *et al*., 2006).

Table 3. Some fish-associated bacterial zoonoses.[a,b,c,d,e,f]

Mechanism of transmission	Disease
Contact-borne	
Mycobacterium marinum, M. fortuitum, M. ulcerans	Fish handler disease, tank granuloma
Streptococcus iniae	Cellulitis, systemic infections
Aeromonas hydrophila, A. sobria, A. caviae	Skin wound infections, systemic infections
Vibrio damselae, V. vulnificus, V. mimicus, V. fluvialis, V. alginolyticus	Skin and wound infections, systemic infections
Edwardsiella tarda	Cellulitis, gastroenteritis, bacteraemia
Erysipelothrix rhusopathiae	Skin infections, systemic infections
Stenotrophomonas maltophilia (?)	Pneumonia, systemic infections
Kluyvera (?)	Gastroenteritis, bacteraemia
Food-borne	
Vibrio parahaemolyticus, V. cholerae	Diarrhoea
Aeromonas hydrophila	Diarrhoea, systemic infections
Salmonella	Diarrhoea, systemic infections
Listeria monocytogenes	Diarrhoea, systemic infections
Clostridium botulinicum, C. perfringens	Botulism, diarrhoea
Plesiomonas shigelloides	Diarrhoea

a. Sarria *et al.* (2001).
b. Looney *et al.* (2009).
c. Lowry and Smith (2007).
d. Iwamoto *et al.* (2010).
e. Boylan (2011).
f. Austin and Austin (2012).

Effects of aquacultural use of antimicrobials on animal and human health

Animal health

The most obvious detrimental effect of extensive use of antimicrobials in aquaculture is selection of fish and shellfish pathogens resistant to multiple antimicrobials which in turn produce difficult or impossible to treat epizootics (L'Abee-Lund and Sørum, 2002; Murray and Peeler, 2005; Toranzo *et al.*, 2005; Asche *et al.*, 2010; Barton and Floysand, 2010; Pulkkinen *et al.*, 2010; Ibieta *et al.*, 2011). The clinical problems generated in veterinary and human medicine by antimicrobial-resistant bacteria are well reviewed (Aarestrup *et al.*, 2000; 2008; Anderson *et al.*, 2003; Angulo *et al.*, 2004; Molbak, 2006; Sapkota *et al.*, 2008; Le Hello *et al.*, 2011; Marshall and Levy, 2011), and fish and shellfish pathogens resistant to multiple antimicrobials used in aquaculture have been described (Austin, 1985; Arthur *et al.*, 2000; Sørum, 2000; 2006; Armstrong *et al.*, 2005; Toranzo *et al.*, 2005). These include *Aeromonas salmonicida*, *A. hydrophila*, *A. caviae*, *A. sobria*, *E. ictaluri*, *E. tarda*, *P. damselae piscicida*, *Vibrio anguillarum, V. salmonicida*, *V. ordalii*, *Flavobacterium psychrophilum*, *Pseudomonas fluorescens, Streptococcus iniae*, *Renibacterium salmonicarum*, *Yersinia ruckeri* and *Piscirickettsia salmonis* (Table 2). In most of them, antimicrobial resistance is mediated by plasmids and MGE, often conjugative, and with potential for HGT (Austin, 1985; Arthur *et al.*, 2000; Rhodes *et al.*, 2000; Sørum, 2000; 2008; Schmidt *et al.*, 2001a; Armstrong *et al.*, 2005; Casas *et al.*, 2005; Toranzo *et al.*, 2005; Erauso *et al.*, 2011). Some of these pathogens, e.g. *Edwarsiella*, *Aeromonas*, and *Streptococcus*, can infect humans and generate antimicrobial-resistant zoonotic infections (Table 3) (Novotny *et al.*, 2004; Toranzo *et al.*, 2005; Lowry and Smith, 2007; Iwamoto *et al.*, 2010; Boylan, 2011; Naviner *et al.*, 2011; Austin and Austin, 2012; Leung *et al.*, 2012). *Kluyvera* spp. and *S. maltophilia* are additional aquatic bacteria related to fish that are emerging as opportunistic human pathogens (Sarria *et al.*, 2001; Looney *et al.*, 2009).

The worldwide occurrence of IncU, pRAS3, pRAS1 and pAr-32 plasmids in *Aeromonas* illustrates the relevance of the widespread dissemination of antimicrobial resistance genes coded by MGE in fish and shellfish pathogens (L'Abee-Lund and Sørum, 2002; Sørum *et al.*, 2003). Similarly, PMQR genes selected in aquatic bacteria as a result of aquacultural antimicrobial use could hypothetically pass by HGT to fish pathogens such as *F. psychrophilum*, *A. salmonicida* and *Y. ruckeri* expressing a mutated GyrA (Oppegaard and Sørum, 1994; Izumi and Aranishi, 2004; Shah *et al.*, 2012a,b). Such an occurrence could increase quinolone resistance and increase quinolone concentrations required to prevent chromosomal mutations to these antimicrobials, and as a result complicate treatment of infections caused by these pathogens (Oppegaard and Sørum, 1994; Izumi and Aranishi, 2004; Strahilevitz *et al.*, 2009; Shah *et al.*, 2012a,b).

While selection of antimicrobial-resistant bacteria in normal intestinal and other flora of fish as a result of aquacultural use of antimicrobials has not been extensively investigated, it is reasonable to suppose that antimicrobial resistance determinants present in normal piscine flora could be the source of resistance

determinants in piscine pathogens analogous to what has been shown to occur in terrestrial animals and human beings (Salyers and Shoemaker, 2006; Navarrete *et al*., 2008; Marshall *et al*., 2009; Martinez *et al*., 2009; Nayak, 2010; Looft *et al*., 2012). Antimicrobial resistance determinants in piscine pathogens could also be acquired from environmental antimicrobial-resistant bacteria that have been selected by residual antimicrobials in water and sediments (Kruse and Sørum, 1994; Davison, 1999; Alonso *et al*., 2001; Fricke *et al*., 2008; Cantón, 2009; Martinez, 2009a; 2012; Allen *et al*., 2010; Fondi and Fani, 2010; Colomer-Lluch *et al*., 2011; Stokes and Gillings, 2011; Dantas and Sommer, 2012). Both of these processes can be stimulated by the presence of antimicrobials in fish tissues and in the environment since (as previously mentioned) many of these antimicrobials are able to fuel HGT and mutagenesis (Aarestrup *et al*., 2000; Beaber *et al*., 2004; Couce and Blazquez, 2009; Kohanski *et al*., 2010; Allen *et al*., 2011; White and McDermott, 2011). Moreover, alterations produced by antimicrobials in the sediments and in the normal flora in the piscine intestinal tract may favour infection by pathogens (Navarrete *et al*., 2008; Nayak, 2010). Excessive and prophylactic use of antimicrobials in aquacultural settings can thus be counterproductive by selecting and favouring untreatable infections with piscine and shellfish pathogens resistant to multiple antimicrobials, and result in the collapse of these activities (Lin, 1989; Holmström *et al*., 2003; León-Muñoz *et al*., 2007; Asche *et al*., 2010; Barton and Floysand, 2010; Ibieta *et al*., 2011; Millanao *et al*., 2011).

Salmon aquaculture requires growth of this anadromous species in both fresh and salt water. This results in differences in normal flora and pathogens in fish and the environment in these two locations, differences which in turn affect the outcome of the selective effects of antimicrobials (Woo *et al*., 2002; Beveridge, 2004; Austin and Austin, 2012). For example, antimicrobials used in freshwater will select for antimicrobial resistance among the freshwater pathogens *F. psychrophilum* and *A. salmonicida* while those used in the marine stages will select among marine pathogens such as *Vibrio* spp. and *P. salmonis* (Woo *et al*., 2002; Beveridge, 2004; Austin and Austin, 2012).

In Chile, the augmented use of antimicrobials that accompanied increases in salmon production coincided with surges in fish mortalities and emergence of new and resistant bacterial pathogens (Asche *et al*., 2010; Ibieta *et al*., 2011; Millanao *et al*., 2011). It was in this period that *S. phocae*, *Rhodococcus qingshengi, F. chilensis and F. araucanum* emerged as potential new salmon pathogens (Valdés *et al*., 2009; Avendaño-Herrera *et al*., 2011; Kämpfer *et al*., 2012). Moreover, approximately 90% of isolates of *F. psychrophilum*, the cause of cold water disease in salmon and trout, were resistant to the three most commonly used antimicrobials (tetracyclines, florfenicol, oxolinic acid) making this disease practically untreatable with them (Henríquez-Núnez *et al*., 2012). In other regions of the world, pathogens of aquacultured fish such as *Edwardsiella ictaluri* and *E. tarda* also display high levels of antimicrobial resistance (Dung *et al*., 2008; Nadirah *et al*., 2012). In Taiwan, the collapse of shrimp aquaculture during the late 1980s resulted from the emergence of multiple-resistant pathogens selected by the injudicious use of antimicrobials (Lin, 1989; Kautsky *et al*., 2000). Preliminary observations suggest that the frequency of detection of antimicrobial resistance genes in aquaculturally-related aquatic bacteria can be correlated with the amounts of antimicrobials used in this activity in Norway and Chile (Shah, 2012).

Human health

There are increasing signs that antimicrobial use in aquaculture may have a long-term and permanent potential to select for antimicrobial-resistant bacteria in the aquatic environment at multiple levels (DePaola *et al*., 1995; Capone *et al*., 1996; Schmidt *et al*., 2001a; Holmström *et al*., 2003; Miranda *et al*., 2003; Sørum, 2006; Miranda and Rojas, 2007; Seyfried *et al*., 2010). This may be particularly relevant to human health in those countries where aquacultural use is heavy, prophylactic and uncontrolled, since bacteria and archaea in the aquatic environment share a large assortment of MGE and antimicrobial resistance genes with a wide range of terrestrial bacteria (Furushita *et al*., 2003; Hastings *et al*., 2004; Furushita and Shiba, 2007; Sobecky and Hazen, 2009; McDaniel *et al*., 2010; Erauso *et al*., 2011; Millanao *et al*., 2011; Taylor *et al*., 2011; Buschmann *et al*., 2012). Indeed, there is strong laboratory and field evidence for readily detectable frequencies of HGT between bacteria in the aquatic environment and human pathogens, as would be expected of genetic exchange communities linked by HGT in spite of the oligotrophy of the aquatic environments (Sandaa *et al*., 1992; Goodman *et al*., 1993; L'Abee-Lund and Sørum, 2002; Furushita and Shiba, 2007; Baquero *et al*., 2008; Guglielmetti *et al*., 2009; Taylor *et al*., 2011; Lupo *et al*., 2012). As a result of HGT, these new genetic entities may be incorporated into the pangenome of terrestrial bacteria including human pathogens, linking the aquatic and terrestrial resistomes and complicating the treatment of human infections (Sandaa *et al*., 1992; Furushita *et al*., 2003; Medini *et al*., 2005; Sørum, 2006; Sobecky and Hazen, 2009; Martinez, 2009a; Fondi and Fani, 2010; Erauso *et al*., 2011; Forsberg *et al*., 2012). The power of HGT to generate genetic diversity from aquatic bacteria is demonstrated by the ability of human intestinal *Bacteroides* to acquire genes needed for degradation of algal polysaccharides

from aquatic bacteria (Hehemann *et al*., 2010; 2012). This gene flow may not be directly from aquatic bacteria to human pathogens but may involve intermediaries such as other environmental bacteria or commensal flora of animals and humans (Roberts, 2009; Skippington and Ragan, 2011). This complex ecological web makes tracking the flow of antimicrobial resistant genes and their history difficult but not impossible (Roberts, 2009; Skippington and Ragan, 2011). While genetic flow between aquatic and terrestrial bacteria might well be restricted by molecular mechanisms against DNA transfer (Thomas and Nielsen, 2005; Martinez *et al*., 2009; Skippington and Ragan, 2011; Wiedenbeck and Cohan, 2011), it still might frequently occur because the strong selective pressure in aquatic sediments contaminated with antimicrobials could overcome these restrictive mechanisms (Hastings *et al*., 2004; Thomas and Nielsen, 2005; Aminov and Mackie, 2007). The potential bidirectional flow of antimicrobial resistance genes between aquatic bacteria and human pathogens increases the danger to human health if this flow results in high risk clones that can disseminate widely among human populations (Woodford *et al*., 2011).

An example of such genetic flow is the occurrence of similar IncU incompatibility group plasmids containing Tn*1721* TetA determinants and integron1 in piscine and human pathogenic *Aeromonas* and in *Escherichia coli* isolated in hospitalized patients (Rhodes *et al*., 2000; 2004; Sørum *et al*., 2003). Sharing of the quinolone resistance genes *qnr*A, *qnrS* and *qnrVC* by aquatic *Shewanella, Photobacterium, Aeromonas* and *Vibrio* (Table 2) with a large array of Gram-negative human pathogens (e.g. *E. coli* and *Klebsiella*) is another example of such gene flows (Poirel *et al*., 2005; 2012; Saga *et al*., 2005; Cattoir *et al*., 2007; 2008; Martínez-Martínez *et al*., 2008; Strahilevitz *et al*., 2009; Xia *et al*., 2010; Hernández *et al*., 2011). We ourselves have found the PMQR gene *aac(6′)-Ib-cr*, commonly found in clinical isolates, in marine bacteria such as *Rhodococcus* spp. (Fig. 3) (Robicsek *et al*., 2006; Buschmann *et al*., 2012; Poirel *et al*., 2012). The current dissemination of CTX-M-type extended-spectrum β-lactamases among enteric pathogens may be a third example of human pathogens probably acquiring antimicrobial resistance genes from aquatic bacteria (Rodriguez *et al*., 2004; Cantón and Coque, 2006; Cantón *et al*., 2012). In this case, it has been postulated that the CTX-M gene was acquired from *Kluyvera*, a genus encountered in the aquatic environment in fish intestines and an opportunistic human pathogen (Tables 2 and 3) (Decousser *et al*., 2001; Sarria *et al*., 2001; Humeniuk *et al*., 2002; Rodriguez *et al*., 2004; Cantón and Coque, 2006; Navarrete *et al*., 2008; Rossolini *et al*., 2008; Cantón *et al*., 2012). Plasmids of the IncA/C incompatibility group harbouring a variety of antimicrobial resistance genetic elements and metal resistance genes have been recently found to be shared by fish pathogens such as *Y. ruckeri*, *Aeromonas, Edwarsiella* (Table 2) and human pathogens such as *Y. pestis*, *Salmonella* and *V. cholerae* (Welch *et al*., 2007; 2009; McIntosh *et al*., 2008; Pan *et al*., 2008; Fricke *et al*., 2009; Call *et al*., 2010; Douard *et al*., 2010; Toleman and Walsh, 2010). It has also been postulated that bacteria such as *Aeromonas* exposed to antimicrobials in an aquatic environment may have facilitated the transfer of the IncA/C plasmids between bacteria of different environments to human pathogens (McIntosh *et al*., 2008; Fricke *et al*., 2009; Parker and Shaw, 2011). A similar role could be played by *Edwardsiella* and *Vibrio* (Pan *et al*., 2008; Welch *et al*., 2009; Leung *et al*., 2012).

The unrestricted transmission of STX/R391 (an ICE able to harbour a multiple antimicrobial resistance integron and to mobilize genomic islands) between aquatic *V. cholerae*, the opportunistic human pathogens *Providencia* and *Proteus*, the fish pathogen, *P. damselae*, and the environmental aquatic, *Shewanella*, underscores the potential for HGT between bacteria from the aquatic environment and human pathogens (Burrus *et al*., 2006; Osorio *et al*., 2008; Wozniak *et al*., 2009; Daccord *et al*., 2010; Wozniak and Waldor, 2010; Toleman and Walsh, 2011; Rodriguez-Blanco *et al*., 2012). ICE elements are genetically related to the IncA/C plasmids with the potential of genetic recombination and interactions between them that facilitate their host range and dissemination (Burrus *et al*., 2006; Osorio *et al*., 2008; Wozniak *et al*., 2009; Daccord *et al*., 2010; Wozniak and Waldor, 2010; Guglielmini *et al*., 2011; Toleman and Walsh, 2011). Ready distribution and transfer of antimicrobial resistance genes between bacteria in the aquatic environment and terrestrial bacteria and human pathogens is further demonstrated by the sharing of *tetG* and *floR* resistance determinants of an antimicrobial-resistance *Salmonella* genomic island 1 (SGI1) between *P. damselae piscicida* and epidemic *S. enterica* serovar Typhimurium DT104, fish-transmitted serovar Paratyphi B, serovar Agona and serovar Albany (Zhao and Aoki, 1992; Kim and Aoki, 1996a; Angulo, 1999; Arcangioli *et al*., 1999; 2000; Bolton *et al*., 1999; Briggs and Fratamico, 1999; Cloeckaert *et al*., 2000; 2001; Boyd *et al*., 2002; 2008; Meunier *et al*., 2002; Doublet *et al*., 2003; Smith, 2008a,b). It is also demonstrated by the suspected potential passage of *tetC* tetracycline island mediated by insertion element IS*cs605* (an insertion element also present in *Helicobacter pylori*) (Lau *et al*., 2008; Roberts, 2009; Roberts and Schwarz, 2009; Sandoz and Rockey, 2010) from aquatic *A. salmonicida* or the opportunistic piscine-originated *Laribacter hongkongensis* to *Chlamydia suis*, a pig pathogen (Lau *et al*., 2008; Roberts, 2009; Roberts and Schwarz, 2009; Sandoz and Rockey, 2010; Roberts *et al*., 2012), and by

the assumed origin in aquaculture of SGI1 variant K in internationally disseminated *S. enterica* serovar Kentucky ST198 resistant to ciprofloxacin (Le Hello *et al*., 2011).

The wide spectrum of potential interactions between these antimicrobial resistance MGEs of aquatic and terrestrial bacteria is further illustrated by a recent report that SGI1 can be mobilized between different bacteria by antimicrobial resistance plasmids of incompatibility group IncA/C found in piscine (*Aeromonas*, *Photobacterium*) and human pathogens (*Salmonella*, *Proteus, Vibrio*) (Douard *et al*., 2010). Undoubtedly, the possibilities of HGT between bacteria of the aquatic environment and human pathogens are increased in settings where injudicious use of antimicrobials in aquaculture facilitates passage of large amounts of antimicrobials into the aquatic environment (Cabello, 2006; Asche *et al*., 2010; Burridge *et al*., 2010; Millanao *et al*., 2011; Buschmann *et al*., 2012). There the antimicrobials can select for antimicrobial-resistant bacteria increasing their numbers, stimulate mutagenesis and HGT, and facilitate dissemination of antimicrobial resistance genes from the aquatic resistome to the terrestrial one (Baya *et al*., 1986; Baquero *et al*., 2008; Cantón, 2009; Couce and Blazquez, 2009; Martinez, 2009a; Forsberg *et al*., 2012; Lupo *et al*., 2012; Tello *et al*., 2012). The use of antimicrobials in aquaculture may also negatively influence human health in areas where the marine aquatic environment is the source of epidemics of shellfish-borne *V. parahaemolyticus* (Cabello *et al*., 2007; García *et al*., 2013). Selecting for antimicrobial-resistant Vibrios in the environment and facilitating the transfer and mutagenesis of its chromosomal *qnr*-like loci and alternative antimicrobial resistance genes to other pathogens (Saga *et al*., 2005; Cabello *et al*., 2007). Contamination of the aquatic environment with human pathogens, as is common in lakes, rivers and the marine litoral in developing countries, will further facilitate genetic flow between aquatic bacteria and these pathogens (Silva *et al*., 1987; Miranda and Zemelman, 2001; Baquero *et al*., 2008; Luna *et al*., 2010; Ribeiro *et al*., 2010; Tello *et al*., 2012). The aquatic environment at aquacultural sites, as suggested by Sørum and Baquero, may constitute a bona fide reactor for facilitating and accelerating evolution towards antimicrobial resistance of a wide range of aquatic and terrestrial bacteria including human pathogens (Sørum, 2006; Baquero *et al*., 2008). In the case of salmonid aquaculture, transport of juvenile fish from hatcheries and lakes to the marine environment will also play a role in disseminating antimicrobial resistant bacteria and genes between these two aquatic environments (Sørum, 2006; Miranda, 2012).

In addition to selection and dissemination of antimicrobial-resistant bacteria, excessive use of antimicrobials in aquaculture can potentially have other detrimental impacts on human health (Austin, 1985; Burka *et al*., 1997; Arthur *et al*., 2000; Haya *et al*., 2001; Cabello, 2006; Hastein *et al*., 2006; Sarmah *et al*., 2006; Sapkota *et al*., 2008; Heuer *et al*., 2009; Rodgers and Furones, 2009; Burridge *et al*., 2010; Abraham, 2011; Naviner *et al*., 2011). Fish products for human consumption can become contaminated with antimicrobial residues at doses higher than Maximum Residue Limits (Silva *et al*., 1987; Cabello, 2006; Silley, 2007; Silbergeld *et al*., 2008; Nogales *et al*., 2011). When such products are eaten, they can potentially alter the human normal intestinal flora, select for antimicrobial-resistant bacteria, and aid infection with human pathogens while further facilitating HGT of antimicrobial resistance (Cabello, 2006; Salyers and Shoemaker, 2006; Silley, 2007; Nisha, 2008; Sapkota *et al*., 2008; Silbergeld *et al*., 2008). Passage of antimicrobials to humans in fish meat may be more common than supposed since regulatory agencies frequently detect antimicrobial residues in fish for human consumption despite the low proportion of aquacultured fish tested for the presence of these drugs (Arthur *et al*., 2000; Sapkota *et al*., 2008; Silbergeld *et al*., 2008; Tacon and Metian, 2008; Rodgers and Furones, 2009; Burridge *et al*., 2010; Love *et al*., 2011). Ingestion of free-ranging (wild) fish, shellfish and crustaceans from areas surrounding aquaculture sites can also result in passage of antimicrobials used in aquaculture to the human intestine since antimicrobials can reach other animals near these sites and remain in their tissues for some period of time (Björklund *et al*., 1990; Sarmah *et al*., 2006; Fortt *et al*., 2007; Sapkota *et al*., 2008). Similarly, antimicrobial-resistant bacteria selected in aquaculture sites can contaminate marketed aquacultural produce (Sarmah *et al*., 2006; Naviner *et al*., 2011; Castillo Neyra *et al*., 2012; Nawaz *et al*., 2012). In addition, there is the reasonable possibility that workers in food mills and aquaculture sites will become exposed to antimicrobials and antimicrobial resistant bacteria by aerosols and by direct contact with medicated food in aquacultural areas where annual usages of antimicrobials run into the metric tons, again shifting the normal flora of skin, intestine and mucosa of these workers towards antimicrobial-resistant bacteria (Burridge *et al*., 2010; Millanao *et al*., 2011; Castillo Neyra *et al*., 2012).

Concluding remarks

Although the available information is partial and fragmented it does not support the hypothesis that the aquatic environment and its bacteria are unique. On the contrary, it strongly suggests that aquaculture, like terrestrial animal farming, is an important source for passage of large amounts of a variety of antimicrobials into the environment. Better information is needed to provide more accurate assessment of the classes and amounts of

antimicrobials used in aquaculture in order to determine their potential impact on the general environment and on animal and public health (Aarestrup *et al*., 2000; 2008; Collignon *et al*., 2009; Heuer *et al*., 2009; Love *et al*., 2011). Despite the lack of accurate information, it is clear that excessive amounts of antimicrobials are used in aquaculture in some countries for both therapeutic and prophylactic purposes (Arthur *et al*., 2000; Armstrong *et al*., 2005; Sapkota *et al*., 2008; Rodgers and Furones, 2009; Asche *et al*., 2010; Barton and Floysand, 2010; Burridge *et al*., 2010; Ndi and Barton, 2012). This veterinary use includes antimicrobials also used clinically in human medicine (Millanao, 2002; Collignon *et al*., 2009; Heuer *et al*., 2009; Millanao *et al*., 2011). Previous experience regarding use of antimicrobials in terrestrial animal husbandry and an analysis of extant information regarding genetic aspects of antimicrobial resistance in aquatic bacteria strongly suggests that antimicrobial use in aquaculture is also likely to select antimicrobial-resistant bacteria (including piscine pathogens) in aquacultural environments (Aarestrup *et al*., 2000; Cabello, 2006; Nikaido, 2009; Levy and Marshall, 2010; Davis *et al*., 2011; Marshall and Levy, 2011; Buschmann *et al*., 2012). Evidence also exists suggesting that the resistome of aquatic bacteria contains novel antimicrobial genetic determinants (Miranda *et al*., 2003; Cattoir *et al*., 2007; 2008). Passage of such antimicrobial resistance determinants from aquatic to terrestrial bacteria will be facilitated by excessive antimicrobial use and the common mobilome of aquatic and terrestrial bacteria (Sørum, 2006; Sobecky and Hazen, 2009; Millanao *et al*., 2011). These novel antimicrobial resistance elements may ultimately reach human pathogens and complicate therapy of infections caused by them (Aarestrup *et al*., 2000; 2008; Miranda *et al*., 2003; Cattoir *et al*., 2007; 2008; Roberts, 2009). The presence of residual antimicrobials in the meat of target and free-ranging species surrounding aquaculture sites and the exposure to antimicrobials of workers that manipulate medicated food is yet another way in which excessive use of antimicrobials in aquaculture may impact human health (Samuelsen *et al*., 1992b; Fortt *et al*., 2007; Sapkota *et al*., 2008). These considerations suggest that excessive aquacultural use of antimicrobials may potentially have major effects on animal and human health as well as on the environment.

The global reach of the problem of antimicrobial resistance indicates that the potential complications of antimicrobial use in aquaculture need to be addressed globally (Angulo, 1999; Anderson *et al*., 2003; Davies, 2009; Martinez *et al*., 2009; Levy and Marshall, 2010). This assessment must include an evaluation of governmental regulations as well as determination of the classes and amounts of antimicrobials used in aquaculture in different countries throughout the world (Davies, 2009; Martinez *et al*., 2009; Burridge *et al*., 2010), and investigation of the reasons aquacultural conglomerates show drastic differences in antimicrobial use in different countries (Grave *et al*., 1999; 2006; Millanao, 2002; Grave and Hansen, 2009; Burridge *et al*., 2010; Millanao *et al*., 2011). Such information is a prerequisite to regulating aquacultural use, especially for those antimicrobials important to human therapeutics. It is also crucial to anticipating potential problems of antimicrobial resistance related to piscine and human health stemming from this use which still goes undetected in a number of countries (Grave *et al*., 1999; 2006; Grave and Hansen, 2009; Asche *et al*., 2010; Burridge *et al*., 2010; Ibieta *et al*., 2011; Millanao *et al*., 2011; Ndi and Barton, 2012). In parallel with this increased assessment of antimicrobial use, there is a need for increased awareness and research focused on the aquatic resistome and the potential passage of genetic elements and antimicrobial resistant determinants from this resistome to the resistomes of fish and human pathogens (Wright *et al*., 2008; Cantón, 2009; Wright, 2010). In this regard, the use of metagenomics with cloning, next generation DNA sequencing and molecular epidemiological tools are already helping to improve definition of the resistome of environmental, animal and human bacteria sharing of antimicrobial resistance genes (Sørum, 2006; Fondi and Fani, 2010; Sommer *et al*., 2010; Kristiansson *et al*., 2011; Sommer and Dantas, 2011; Forsberg *et al*., 2012).

Regulation of antimicrobial use in farmed animals in Europe and in salmon farms in Norway has demonstrated that reducing the use of antimicrobials is not incompatible with economically feasible animal farming (Markestad and Grave, 1997; Aarestrup *et al*., 2000; 2008; Wegener, 2003; Sørum, 2006; Midtlyng *et al*., 2011; White and McDermott, 2011). There is thus a critical need to educate all stakeholders (including aquacultural corporations) to understand that sacrificing fish hygiene and well-being for short-term economic gains is not a winning strategy, and that appropriate use of prebiotics, probiotics and vaccines can replace excessive use of antimicrobials (Markestad and Grave, 1997; Bravo and Midtlyng, 2007; Defoirdt *et al*., 2007). The continuous growth of aquaculture and the potential increase of fish diseases generated by global warming and globalization increases the urgency of coupling these approaches so that all can reap maximal benefits from antimicrobial use while avoiding the negative effects of their excessive use on the environment and on animal and human health (Sapkota *et al*., 2008; Sørum, 2008; Asche, 2009; Asche *et al*., 2010).

Acknowledgements

This work was supported by a grant from the Lenfest Ocean Program/Pew Charitable Trusts to F. C. C. and A. B. and by a

fellowship from the John Simon Guggenheim Foundation to F. C. C. that allowed him to begin to study antimicrobial use in aquaculture. A stimulus for writing this review was an invitation to F. C. C. to participate in a symposium, 'The Effects of Semi-Intensive Aquaculture on Biodiversity in Nearshore and Inland Waters', at the Annual Meeting in Seattle of the American Fisheries Society, September 2011. This symposium was supported by USAID CA/LWA No. EPP-A-00–06-00012-00 through the AquaFish Collaborative Research Support Program (CRSP) and also by participating institutions. We thank Dr Les E. Burridge, Fisheries and Oceans Canada, St Andrews Biological Station, St Andrews, New Brunswick, Canada; Dr James S. Diana, School of Natural Resources and Environment, University of Michigan, Ann Arbor, MI, USA; and Dr Karen Levy, Dept. of Environmental Health, Rollins School of Public Health, Emory University, Atlanta, GA, USA, for valuable suggestions in writing the review. We also acknowledge the important suggestions of an anonymous reviewer. H. D. and A. M. thank the support of Laboratorio GSK Chile. We thank Dr S. B. Levy and L. M. McMurry, Tufts Medical School, Boston, MA, USA, for *E. coli* DH5α pET*tetB1* and *S. aureus* pT18; Dr T. Kitten, The Philips Institute of Oral and Craniofacial Molecular Biology, VCU School of Dentistry, Richmond, VA, USA, for *E. coli* DH10B pJFP76; and Dr G. A. Jacoby, Lahey Clinic, Burlington, MA, USA, for *E. coli* J53 pMG298. We thank Dr María Luisa Rioseco, Hospital Regional, Puerto Montt, Chile, for the clinical isolates of urinary *E. coli* strains. We also thank Mrs Harriett V. Harrison for her invaluable help during preparation of the manuscript.

References

Aarestrup, F.M., Seyfarth, A.-M., Emborg, H.-D., Bager, F., Pedersen, K., and Jorsal, S.E. (2000) Antibiotic use in food-animal production in Denmark. *APUA Newsletter* **18:** 4–5.

Aarestrup, F.M., Wegener, H.C., and Collignon, P. (2008) Resistance in bacteria of the food chain: epidemiology and control strategies. *Expert Rev Anti Infect Ther* **6:** 733–750.

Abraham, T.J. (2011) Food safety hazards related to emerging antibiotic resistant bacteria in cultured freshwater fishes of Kolkata, India. *Adv J Food Sci Technol* **3:** 69–72.

Akinbowale, O.L., Peng, H., and Barton, M.D. (2006) Antimicrobial resistance in bacteria isolated from aquaculture sources in Australia. *J Appl Microbiol* **100:** 1103–1113.

Akinbowale, O.L., Peng, H., Grant, P., and Barton, M.D. (2007) Antibiotic and heavy metal resistance in motile aeromonads and pseudomonads from rainbow trout *(Oncorhynchus mykiss)* farms in Australia. *Int J Antimicrob Agents* **30:** 177–182.

Alcaide, E., Blasco, M.D., and Esteve, C. (2005) Occurrence of drug-resistant bacteria in two European eel farms. *Appl Environ Microbiol* **71:** 3348–3350.

Alekshun, M.N., and Levy, S.B. (2007) Molecular mechanisms of antibacterial multidrug resistance. *Cell* **128:** 1037–1050.

Allen, H.K., Donato, J., Wang, H.H., Cloud-Hansen, K.A., Davies, J., and Handelsman, J. (2010) Call of the wild: antibiotic resistance genes in natural environments. *Nat Rev Microbiol* **8:** 251–259.

Allen, H.K., Looft, T., Bayles, D.O., Humphrey, S., Levine, U.Y., Alt, D., and Stanton, T.B. (2011)) Antibiotics in feed induce prophages in swine fecal microbiomes. *mBio* **2:** e00260–11.

Alonso, A., Sanchez, P., and Martinez, J.L. (2001) Environmental selection of antibiotic resistance genes. *Environ Microbiol* **3:** 1–9.

American Academy of Microbiology (2009) *Antibiotic Resistance: An Ecological Perspective on an Old Problem*. Washington, DC, USA: American Academy of Microbiology.

Aminov, R.I., and Mackie, R.I. (2007) Evolution and ecology of antibiotic resistance genes. *FEMS Microbiol Lett* **271:** 147–161.

Anderson, A.D., Nelson, J.M., Rossiter, S., and Angulo, F.J. (2003) Public health consequences of use of antimicrobial agents in food animals in the United States. *Microb Drug Resist* **9:** 373–379.

Andersson, D.I., and Hughes, D. (2011) Persistence of antibiotic resistance in bacterial populations. *FEMS Microbiol Rev* **35:** 901–911.

Angulo, F. (1999) *Use of Antimicrobial Agents in Aquaculture: Potential for Public Health Impact*. Atlanta, GA, USA: Centers for Disease Control and Prevention, Department of Health and Human Services. [WWW document]. URL http://www.fda.gov/ohrms/dockets/dailys/00/apr00/041100/c000019.pdf.

Angulo, F.J., Nargund, V.N., and Chiller, T.C. (2004) Evidence of an association between use of anti-microbial agents in food animals and anti-microbial resistance among bacteria isolated from humans and the human health consequences of such resistance. *J Vet Med B Infect Dis Vet Public Health* **51:** 374–379.

Aoki, T., Satoh, T., and Kitao, T. (1987) New tetracycline resistance determinant on R plasmids from *Vibrio anguillarum*. *Antimicrob Agents Chemother* **31:** 1446–1449.

Arcangioli, M.A., Leroy-Setrin, S., Martel, J.L., and Chaslus-Dancla, E. (1999) A new chloramphenicol and florfenicol resistance gene flanked by two integron structures in *Salmonella typhimurium* DT104. *FEMS Microbiol Lett* **174:** 327–332.

Arcangioli, M.A., Leroy-Setrin, S., Martel, J.L., and Chaslus-Dancla, E. (2000) Evolution of chloramphenicol resistance, with emergence of cross-resistance to florfenicol, in bovine *Salmonella typhimurium* strains implicates definitive phage type (DT) 104. *J Med Microbiol* **49:** 103–110.

Armstrong, S.M., Hargrave, B.T., and Haya, K. (2005) Antibiotic use in finfish aquaculture: modes of action, environmental fate, and microbial resistance. *Hdbk Environ Chem* **5M:** 341–357.

Arthur, J.R., Lavilla-Pitogo, C.R., and Subasinghe, R.P. (2000) *Use of Chemicals in Aquaculture in Asia. Proceedings of the Meeting on the Use of Chemicals in Aquaculture in Asia. Tighauan, Iloilo, Philippines*. Tigbauan, Philippines: Southeast Asian Fisheries.

Asche, F. (2009) Farming the sea. *Mar Resource Econ* **23:** 507–527.

Asche, F., Roll, K.H., and Tveterås, S. (2008) Future trends in aquaculture: productivity growth and increased production.

In *Aquaculture in the Ecosystem*. Holmer, M., Black, K., Duarte, C.M., Marbà, N., and Karakassis, I. (eds). London, UK: Springer, pp. 271–292.

Asche, F., Hansen, H., Tveterås, R., and Tveterås, S. (2010) The salmon disease crisis in Chile. *Mar Resource Econ* **24:** 405–411.

Austin, B. (1985) Antibiotic pollution from fish farms: effects on aquatic microflora. *Microbiol Sci* **2:** 113–117.

Austin, B., and Austin, D.A. (2012) *Bacterial Fish Pathogens: Disease of Farmed and Wild Fish*, 5th edn. London, UK: Springer.

Avendaño-Herrera, R., Balboa, S., Doce, A., Ilardi, P., Lovera, P., Toranzo, A.E., and Romalde, J.L. (2011) Pseudo-membranes on internal organs associated with *Rhodococcus gingshengii* infection in Atlantic salmon (*Salmo salar*). *Vet Microbiol* **147:** 200–204.

Baharoglu, Z., and Mazel, D. (2011) *Vibrio cholerae* triggers SOS and mutagenesis in response to a wide range of antibiotics: a route towards multiresistance. *Antimicrob Agents Chemother* **55:** 2438–2441.

Baharoglu, Z., Bikard, D., and Mazel, D. (2010) Conjugative DNA transfer induces the bacterial SOS response and promotes antibiotic resistance development through integron activation. *PLoS Genet* **6:** e1001165.

Baharoglu, Z., Krin, E., and Mazel, D. (2012) Connecting environment and genome plasticity in the characterization of transformation-induced SOS regulation and carbon catabolite control of the *Vibrio cholerae* integron integrase. *J Bacteriol* **194:** 1659–1667.

Baker-Austin, C., McArthur, J.V., Lindell, A.H., Wright, M.S., Tuckfield, R.C., Gooch, J., *et al*. (2009) Multi-site analysis reveals widespread antibiotic resistance in the marine pathogen *Vibrio vulnificus*. *Microb Ecol* **57:** 151–159.

Baquero, F., Martinez, J.L., and Cantón, R. (2008) Antibiotics and antibiotic resistance in water environments. *Curr Opin Biotechnol* **19:** 260–265.

Baquero, F., Alvarez-Ortega, C., and Martinez, J.L. (2009) Ecology and evolution of antibiotic resistance. *Environ Microbiol Rep* **1:** 260–265.

Barnes, A.C., Hastings, T.S., and Amyes, S.G.B. (1995) Aquaculture antibacterials are antagonized by seawater cations. *J Fish Dis* **18:** 463–465.

Barrientos, M. (2006) *Estudio cualitativo y cuantitativo de las quinolonas y fluoroquinolonas importadas y autorizadas para uso y disposición en medicina y en veterinaria en Chile, en el período 2002-2005. Consideraciones sobre su impacto para la salud pública y el medio ambiente. [Qualitative and Quantitative Study of Quinolones and Fluorquinolones Imported and Authorized for Veterinary and Medical Use in Chile, 2002-2005. Considerations of Its Impact on Public Health and the Environment]*. Valdivia, Chile: Universidad Austral de Chile. [WWW document]. URL http://cybertesis.uach.cl/tesis/uach/2006/fcb2751e/doc/fcb2751e.pdf.

Barton, B.A., and Iwama, G.K. (1991) Physiological changes in fish from stress in aquaculture with emphasis on the response and effects of corticosteroids. *Ann Rev Fish Dis* **1:** 3–26.

Barton, J.R., and Floysand, A. (2010) The political ecology of Chilean salmon aquaculture, 1982-2010: a trajectory from economic development to global sustainability. *Glob Environ Change* **20:** 739–752.

Baya, A.M., Brayton, P.R., Brown, V.L., Grimes, D.J., Russek-Cohen, E., and Colwell, R.R. (1986) Coincident plasmids and antimicrobial resistance in marine bacteria isolated from polluted and unpolluted Atlantic Ocean samples. *Appl Environ Microbiol* **51:** 1285–1292.

Beaber, J.W., Hochhut, B., and Waldor, M.K. (2004) SOS response promotes horizontal dissemination of antibiotic resistance genes. *Nature* **427:** 72–74.

Beveridge, M. (2004) *Cage Aquaculture*, 3rd edn., Oxford, UK: Blackwell Publishing.

Biers, E.J., Sun, S., and Howard, E.C. (2009) Prokaryotic genomes and diversity in surface ocean waters: interrogating the global ocean sampling metagenome. *Appl Environ Microbiol* **75:** 2221–2229.

Bissett, A., Bowman, J., and Burke, C. (2006) Bacterial diversity in organically-enriched fish farm sediments. *FEMS Microbiol Ecol* **55:** 48–56.

Björklund, H., Bondestam, J., and Bylund, G. (1990) Residues of oxytetracycline in wild fish and sediments from fish farms. *Aquaculture* **86:** 359–367.

Björklund, H.V., Råbergh, C.M.I., and Bylund, G. (1991) Residues of oxolinic acid and oxytetracycline in fish and sediments from fish farms. *Aquaculture* **97:** 85–96.

Blazquez, J., Couce, A., Rodriguez-Beltran, J., and Rodriguez-Rojas, A. (2012) Antimicrobials as promoters of genetic variation. *Curr Opin Microbiol* **15:** 561–569.

Bolton, L.F., Kelley, L.C., Lee, M.D., Fedorka-Cray, P.J., and Maurer, J.J. (1999) Detection of multidrug-resistant *Salmonella enterica* serotype Typhimurium DT104 based on a gene which confers cross-resistance to florfenicol and chloramphenicol. *J Clin Microbiol* **37:** 1348–1351.

Boucher, Y., Labbate, M., Koenig, J.E., and Stokes, H.W. (2007) Integrons: mobilizable platforms that promote genetic diversity in bacteria. *Trends Microbiol* **15:** 301–309.

Boxall, A.B., Fogg, L.A., Blackwell, P.A., Kay, P., Pemberton, E.J., and Croxford, A. (2004) Veterinary medicines in the environment. *Rev Environ Contam Toxicol* **180:** 1–91.

Boyd, D., Cloeckaert, A., Chaslus-Dancla, E., and Mulvey, M.R. (2002) Characterization of variant *Salmonella* genomic island 1 multidrug resistance regions from serovars Typhimurium DT104 and Agona. *Antimicrob Agents Chemother* **46:** 1714–1722.

Boyd, D.A., Shi, X., Hu, Q.H., Ng, L.K., Doublet, B., Cloeckaert, A., and Mulvey, M.R. (2008) *Salmonella* genomic island 1 (SGI1), variant SGI1-I, and new variant SGI1-O in *Proteus mirabilis* clinical and food isolates from China. *Antimicrob Agents Chemother* **52:** 340–344.

Boylan, S. (2011) Zoonoses associated with fish. *Vet Clin North Am Exot Anim Pract* **14:** 427–438.

Bravo, S., and Midtlyng, P.J. (2007) The use of fish vaccines in the Chilean salmon industry 1999-2003. *Aquaculture* **270:** 36–42.

Briggs, C.E., and Fratamico, P.M. (1999) Molecular characterization of an antibiotic resistance gene cluster of *Salmonella typhimurium* DT104. *Antimicrob Agents Chemother* **43:** 846–849.

Burka, J.F., Hammell, K.L., Horsberg, T.E., Johnson, G.R., Rainnie, D.J., and Speare, D.J. (1997) Drugs in salmonid

aquaculture – a review. *J Vet Pharmacol Ther* **20:** 333–349.

Burridge, L., Weis, J.S., Cabello, F., Pizarro, J., and Bostick, K. (2010) Chemical use in salmon aquaculture: a review of current practices and possible environmental effects. *Aquaculture* **306:** 7–23.

Burrus, V., Quezada-Calvillo, R., Marrero, J., and Waldor, M.K. (2006) SXT-related integrating conjugative element in New World *Vibrio cholerae*. *Appl Environ Microbiol* **72:** 3054–3057.

Buschmann, A.H., Cabello, F., Young, K., Carvajal, J., Varela, D.A., and Henríquez, L. (2009) Salmon aquaculture and coastal ecosystem health in Chile: analysis of regulations, environmental impacts and bioremediation systems. *Ocean Coast Manag* **52:** 243–249.

Buschmann, A.H., Tomova, A., López, A., Maldonado, M.A., Henríquez, L.A., Ivanova, L., *et al*. (2012) Salmon aquaculture and antimicrobial resistance in the marine environment. *PLoS ONE* **7:** e42724.

Bush, K., Courvalin, P., Dantas, G., Davies, J., Eisenstein, B., Huovinen, P., *et al*. (2011) Tackling antibiotic resistance. *Nat Rev Microbiol* **9:** 894–896.

Bushman, F. (2002) *Lateral DNA Transfer. Mechanisms and Consequences*. Cold Spring Harbor, New York, USA: Cold Spring Harbor Laboratory Press.

Cabello, F.C. (2006) Heavy use of prophylactic antibiotics in aquaculture: a growing problem for human and animal health and for the environment. *Environ Microbiol* **8:** 1137–1144.

Cabello, F.C. (2009) Aquaculture and florfenicol resistance in *Salmonella enterica* serovar Typhimurium DT104. *Emerg Infect Dis* **15:** 623–624.

Cabello, F.C., Espejo, R.T., Hernandez, M.C., Rioseco, M.L., Ulloa, J., and Vergara, J.A. (2007) *Vibrio parahaemolyticus* O3:K6 epidemic diarrhea, Chile, 2005. *Emerg Infect Dis* **13:** 655–656.

Call, D.R., Singer, R.S., Meng, D., Broschat, S.L., Orfe, L.H., Anderson, J.M., *et al*. (2010) bla_{CMY-2}-positive IncA/C plasmids from *Escherichia coli* and *Salmonella enterica* are a distinct component of a larger lineage of plasmids. *Antimicrob Agents Chemother* **54:** 590–596.

Cambray, G., Guerout, A.-M., and Mazel, D. (2010) Integrons. *Annu Rev Genet* **44:** 141–165.

Cambray, G., Sanchez-Alberola, N., Campoy, S., Guerin, É., Da Re, S., González-Zorn, B., *et al*. (2011) Prevalence of SOS-mediated control of integron integrase expression as an adaptive trait of chromosomal and mobile integrons. *Mobile DNA* **2:** 6.

Cantón, R. (2009) Antibiotic resistance genes from the environment: a perspective through newly identified antibiotic resistance mechanisms in the clinical setting. *Clin Microbiol Infect* **15** (Suppl. 1): 20–25.

Cantón, R., and Coque, T.M. (2006) The CTX-M β-lactamase pandemic. *Curr Opin Microbiol* **9:** 466–475.

Cantón, R., González-Alba, J.-M., and Galán, J.C. (2012) CTX-M enzymes: origin and diffusion. *Front Microbiol* **3:** Article 110.

Capone, D.G., Weston, D.P., Miller, V., and Shoemaker, C. (1996) Antibacterial residues in marine sediments and invertebrates following chemotherapy in aquaculture. *Aquaculture* **145:** 55–75.

Casas, C., Anderson, E.C., Ojo, K.K., Keith, I., Whelan, D., Rainnie, D., and Roberts, M.C. (2005) Characterization of pRAS1-like plasmids from atypical North American psychrophilic *Aeromonas salmonicida*. *FEMS Microbiol Lett* **242:** 59–63.

Castillo Neyra, R., Vegosen, L., Davis, M.F., Price, L., and Silbergeld, E.K. (2012) Antimicrobial-resistant bacteria: an unrecognized work-related risk in food animal production. *Saf Health Work* **3:** 85–91.

Cattoir, V., Poirel, L., Mazel, D., Soussy, C.J., and Nordmann, P. (2007) *Vibrio splendidus* as the source of plasmid-mediated QnrS-like quinolone resistance determinants. *Antimicrob Agents Chemother* **51:** 2650–2651.

Cattoir, V., Poirel, L., Aubert, C., Soussy, C.J., and Nordmann, P. (2008) Unexpected occurrence of plasmid-mediated quinolone resistance determinants in environmental *Aeromonas* spp. *Emerg Infect Dis* **14:** 231–237.

Chelossi, E., Vezzulli, L., Milano, A., Branzoni, M., Fabiano, M., Riccardi, G., and Banat, I.M. (2003) Antibiotic resistance of benthic bacteria in fish-farm and control sediments of the Western Mediterranean. *Aquaculture* **219:** 83–97.

Chopin, T., Robinson, S.M.C., Troell, M., Neori, A., Buschmann, A., and Fang, J.G. (2008) Ecological engineering: multi-trophic integration for sustainable marine aquaculture. In *Encyclopedia of Ecology*. Jørgensen, S.E., and Fath, B. (eds). Amsterdam, the Netherlands: Elsevier, pp. 2463–2475.

Christensen, A.M., Ingerslev, F., and Baun, A. (2006) Exotoxicity of mixtures of antibiotics used in aquacultures. *Environ Toxicol Chem* **25:** 2208–2215.

Cloeckaert, A., Boumedine, K.S., Flaujac, G., Imberechts, H., D'Hooghe, I., and Chaslus-Dancla, E. (2000) Occurrence of a *Salmonella enterica* serovar Typhimurium DT104-like antibiotic resistance gene cluster including the *floR* gene in *S. enterica* serovar Agona. *Antimicrob Agents Chemother* **44:** 1359–1361.

Cloeckaert, A., Baucheron, S., and Chaslus-Dancla, E. (2001) Nonenzymatic chloramphenicol resistance mediated by IncC plasmid R55 is encoded by a *floR* gene variant. *Antimicrob Agents Chemother* **45:** 2381–2382.

Cole, D.W., Cole, R., Gaydos, S.J., Gray, J., Hyland, G., Jacques, M.L., *et al*. (2009) Aquaculture: environmental, toxicological, and health issues. *Int J Hyg Environ Health* **212:** 369–377.

Collignon, P., Powers, J.H., Chiller, T.M., Aidara-Kane, A., and Aarestrup, F.M. (2009) World Health Organization ranking of antimicrobials according to their importance in human medicine: a critical step for developing risk management strategies for the use of antimicrobials in food production animals. *Clin Infect Dis* **49:** 132–141.

Colomer-Lluch, M., Jofre, J., and Muniesa, M. (2011) Antibiotic resistance genes in the bacteriophage DNA fraction of environmental samples. *PLoS ONE* **6:** e17549.

Cordova-Kreylos, A.L., and Scow, K.M. (2007) Effects of ciprofloxacin on salt marsh sediment microbial communities. *ISME J* **1:** 585–595.

Costa-Pierce, B.A. (2003) Use of ecosystems science in ecological aquaculture. *Bull Aquacul Assoc Canada* **103-2:** 32–40.

Costa-Pierce, B.A. (2010) Sustainable ecological aquaculture systems: the need for a new social contract for aquaculture development. *Mar Technol Soc J* **44:** 88–112.

Couce, A., and Blazquez, J. (2009) Side effects of antibiotics on genetic variability. *FEMS Microbiol Rev* **33:** 531–538.

Coyne, R., Hihey, M., and Smith, P. (1997) Transient presence of oxytetracycline in blue mussels (*Mytilus edulis*) following its therapeutic use at a marine Atlantic salmon farm. *Aquaculture* **149:** 175–181.

Coyne, R., Smith, P., and Moriarty, C. (2001) The fate of oxytetracycline in the marine environment of a salmon cage farm. *Mar Environ Health* **3:** 1–24.

Crossman, L.C., Gould, V.C., Dow, J.M., Vernikos, G.S., Okazaki, A., Sebaihia, M., *et al*. (2008) The complete genome, comparative and functional analysis of *Stenotrophomonas maltophilia* reveals an organism heavily shielded by drug resistance determinants. *Genome Biol* **9:** R74.

D'Costa, V.M., King, C.E., Kalan, L., Morar, M., Sung, W.W., Schwarz, C., *et al*. (2011) Antibiotic resistance is ancient. *Nature* **477:** 457–461.

Daccord, A., Ceccarelli, D., and Burrus, V. (2010) Integrating conjugative elements of the SXT/R391 family trigger the excision and drive the mobilization of a new class of *Vibrio* genomic islands. *Mol Microbiol* **78:** 576–588.

Dang, H., Zhang, X., Song, L., Chang, Y., and Yang, G. (2007) Molecular determination of oxytetracycline-resistant bacteria and their resistance genes from mariculture environments of China. *J Appl Microbiol* **103:** 2580–2592.

Dang, S.T., Petersen, A., Van Truong, D., Chu, H.T., and Dalsgaard, A. (2011) Impact of medicated feed on the development of antimicrobial resistance in bacteria at integrated pig-fish farms in Vietnam. *Appl Environ Microbiol* **77:** 4494–4498.

Dantas, G., and Sommer, M.O. (2012) Context matters – the complex interplay between resistome genotypes and resistance phenotypes. *Curr Opin Microbiol* **15:** 577–582.

Datta, N., and Hughes, V.M. (1983) Plasmids of the same Inc groups in *Enterobacteria* before and after the medical use of antibiotics. *Nature* **306:** 616–617.

Davies, J. (2009) Antibiotic resistance and the future of antibiotics. In *Microbial Evolution and Co-Adaptation. A Tribute to the Life and Scientific Legacies of Joshua Lederberg*. Relman, D.A., Hamburg, M.A., Choffnes, E.R., and Mack, A. (eds). Washington, DC, USA: The National Academies Press, pp. 160–172.

Davies, J., and Davies, D. (2010) Origins and evolution of antibiotic resistance. *Microbiol Mol Biol Rev* **74:** 417–433.

Davis, M.F., Price, L.B., Liu, C.M., and Silbergeld, E.K. (2011) An ecological perspective on U.S. industrial poultry production: the role of anthropogenic ecosystems on the emergence of drug-resistant bacteria from agricultural environments. *Curr Opin Microbiol* **14:** 244–250.

Davison, J. (1999) Genetic exchange between bacteria in the environment. *Plasmid* **42:** 73–91.

Decousser, J.W., Poirel, L., and Nordmann, P. (2001) Characterization of a chromosomally encoded extended-spectrum class A β-lactamase from *Kluyvera cryocrescens*. *Antimicrob Agents Chemother* **45:** 3595–3598.

Defoirdt, T., Boon, N., Sorgeloos, P., Verstraete, W., and Bossier, P. (2007) Alternatives to antibiotics to control bacterial infections: luminescent vibriosis in aquaculture as an example. *Trends Biotechnol* **25:** 472–479.

DePaola, A., Peeler, J.T., and Rodrick, G.E. (1995) Effect of oxytetracycline-medicated feed on antibiotic resistance of gram-negative bacteria in catfish ponds. *Appl Environ Microbiol* **61:** 2335–2340.

Diana, J.S. (2009) Aquaculture production and biodiversity conservation. *BioScience* **59:** 27–38.

Ding, C., and He, J. (2010) Effect of antibiotics in the environment on microbial populations. *Appl Microbiol Biotechnol* **87:** 925–941.

Domingues, S., Harms, K., Fricke, W.F., Johnsen, P.J., da Silva, G.J., and Nielsen, K.M. (2012) Natural transformation facilitates transfer of transposons, integrons and gene cassettes between bacterial species. *PLoS Pathog* **8:** e1002837.

Dorr, T., Lewis, K., and Vulic, M. (2009) SOS response induces persistence to fluoroquinolones in *Escherichia coli*. *PLoS Genet* **5:** e1000760.

Douard, G., Praud, K., Cloeckaert, A., and Doublet, B. (2010) The *Salmonella* genomic island 1 is specifically mobilized in trans by the IncA/C multidrug resistance plasmid family. *PLoS ONE* **5:** e15302.

Doublet, B., Lailler, R., Meunier, D., Brisabois, A., Boyd, D., Mulvey, M.R., *et al*. (2003) Variant *Salmonella* genomic island 1 antibiotic resistance gene cluster in *Salmonella enterica* serovar Albany. *Emerg Infect Dis* **9:** 585–591.

Dung, T.T., Haesebrouck, F., Tuan, N.A., Sorgeloos, P., Baele, M., and Decostere, A. (2008) Antimicrobial susceptibility pattern of *Edwardsiella ictaluri* isolates from natural outbreaks of bacillary necrosis of *Pangasianodon hypophthalmus* in Vietnam. *Microb Drug Resist* **14:** 311–316.

Edlund, A., Soule, T., Sjoling, S., and Jansson, J.K. (2006) Microbial community structure in polluted Baltic Sea sediments. *Environ Microbiol* **8:** 223–232.

Erauso, G., Lakhal, F., Bidault-Toffin, A., Le Chevalier, P., Bouloc, P., Paillard, C., and Jacq, A. (2011) Evidence for the role of horizontal transfer in generating pVT1, a large mosaic conjugative plasmid from the clam pathogen, *Vibrio tapetis*. *PLoS ONE* **6:** e16759.

FAO (2010) *FAO Yearbook*. Fishery and Aquaculture Statistics. Table A4 (p. 28). Rome, Italy: Food and Agriculture Organization of the United Nations, 2012. [WWW document]. URL ftp://ftp.fao.org/FI/CDrom/CD_yearbook_2010/booklet/ba0058t.pdf.

Fernández-Alarcón, C., Miranda, C.D., Singer, R.S., López, Y., Rojas, R., Bello, H., *et al*. (2010) Detection of the *floR* gene in a diversity of florfenicol resistant Gram-negative bacilli from freshwater salmon farms in Chile. *Zoonoses Public Health* **57:** 181–188.

Fondi, M., and Fani, R. (2010) The horizontal flow of the plasmid resistome: clues from inter-generic similarity networks. *Environ Microbiol* **12:** 3228–3242.

Forsberg, K.J., Reyes, A., Wang, B., Selleck, E.M., Sommer, M.O.A., and Dantas, G. (2012) The shared antibiotic resistome of soil bacteria and human pathogens. *Science* **337:** 1107–1111.

Fortt, A.Z., Cabello, F.C., and Buschmann, A.R. (2007) Residuos de tetraciclina y quinolonas en peces silvestres

en una zona costera donde se desarrolla la acuicultura del salmón en Chile [Residues of tetracycline and quinolones in wild fish living around a salmon aquaculture center in Chile]. *Rev Chil Infect* **24:** 14–18.

Fricke, W.F., Wright, M.S., Lindell, A.H., Harkins, D.M., Baker-Austin, C., Ravel, J., and Stepanauskas, R. (2008) Insights into the environmental resistance gene pool from the genome sequence of the multidrug-resistant environmental isolate *Escherichia coli* SMS-3-5. *J Bacteriol* **190:** 6779–6794.

Fricke, W.F., Welch, T.J., McDermott, P.F., Mammel, M.K., LeClerc, J.E., White, D.G., *et al*. (2009) Comparative genomics of the IncA/C multidrug resistance plasmid family. *J Bacteriol* **191:** 4750–4757.

Frost, L.S., Leplae, R., Summers, A.O., and Toussaint, A. (2005) Mobile genetic elements: the agents of open source evolution. *Nat Rev Microbiol* **3:** 722–732.

Furushita, M., and Shiba, T. (2007) Distribution and horizontal transfer of antibiotic resistance gene in marine environments. *J Natl Fish Univ* **56:** 85–89.

Furushita, M., Shiba, T., Maeda, T., Yahata, M., Kaneoka, A., Takahashi, Y., *et al*. (2003) Similarity of tetracycline resistance genes isolated from fish farm bacteria to those from clinical isolates. *Appl Environ Microbiol* **69:** 5336–5342.

García, K., Bastías, R., Higuera, G., Torres, R., Mellado, A., Uribe, P., and Espejo, R.T. (2013) Rise and fall of pandemic *Vibrio parahaemolyticus* serotype O3:K6 in southern Chile. *Environ Microbiol* **15:** 527–534.

Gaze, W.H., Zhang, L., Abdouslam, N.A., Hawkey, P.M., Calvo-Bado, L., Royle, J., *et al*. (2011) Impacts of anthropogenic activity on the ecology of class 1 integrons and integron-associated genes in the environment. *ISME J* **5:** 1253–1261.

Gillings, M., Boucher, Y., Labbate, M., Holmes, A., Krishnan, S., Holley, M., and Stokes, H.W. (2008) The evolution of class 1 integrons and the rise of antibiotic resistance. *J Bacteriol* **190:** 5095–5100.

Giraud, F., Douet, D.-G., Le Bris, H., Bouju-Albert, A., Donnay-Moreno, C., Thorin, C., and Pouliquen, H. (2006) Survey of antibiotic resistance in an integrated marine aquaculture system under oxolinic acid treatment. *FEMS Microbiol Ecol* **55:** 439–448.

Godoy, M.G., Aedo, A., Kibenge, M.J., Groman, D.B., Yason, C.V., Grothusen, H., *et al*. (2008) First detection, isolation and molecular characterization of infectious salmon anaemia virus associated with clinical disease in farmed Atlantic salmon (*Salmo salar*) in Chile. *BMC Vet Res* **4:** 28.

Gómez, C. (2009) *Estudio cualitativo y cuantitativo de las tetraciclinas y fenicoles importadas y autorizadas para uso y disposición en medicina y en veterinaria en Chile, en el período 2002-2005. Consideraciones sobre su impacto para la salud pública y el medio ambiente. [Qualitative and Quantitative Study of Tetracyclines and Phenicols Imported and Authorized for Veterinary and Medical Use in Chile, 2000-2007. Considerations of Its Impact on Public Health and the Environment]*. Valdivia, Chile: Tesis de Grado, Escuela de Química y Farmacia, Universidad Austral de Chile. [WWW document]. URL http://cybertesis.uach.cl/tesis/uach/2009/fcg633e/doc/fcg633e.pdf.

Gonçalves Ferreira, C.S., Nunes, A.N., de Melo Henriques, J.M., and Guilhermino, L. (2007) Acute toxicity of oxytetracycline and florfenicol to the microalgae *Tetraselmis chuii* and to the crustacean *Artemia parthenogenetica*. *Ecotoxicol Environ Saf* **67:** 452–458.

Goodman, A.E., Hild, E., Marshall, K.C., and Hermansson, M. (1993) Conjugative plasmid transfer between bacteria under simulated marine oligotrophic conditions. *Appl Environ Microbiol* **59:** 1035–1040.

Gordon, L., Giraud, E., Ganiere, J.P., Armand, F., Bouju-Albert, A., de la Cotte, N., *et al*. (2007) Antimicrobial resistance survey in a river receiving effluents from freshwater fish farms. *J Appl Microbiol* **102:** 1167–1176.

Gordon, L., Cloeckaert, A., Doublet, B., Schwarz, S., Bouju-Albert, A., Ganiere, J.P., *et al*. (2008) Complete sequence of the *floR*-carrying multiresistance plasmid pAB5S9 from freshwater *Aeromonas bestiarum*. *J Antimicrob Chemother* **62:** 65–71.

Grave, K., and Hansen, M.K. (2009) Previous and current trends in the usage of antimicrobial drugs in Norwegian aquaculture. [WWW document]. URL http://www.fhi.no/dokumenter/ef83612534.pdf.

Grave, K., Lingaas, E., Bangen, M., and Ronning, M. (1999) Surveillance of the overall consumption of antibacterial drugs in humans, domestic animals and farmed fish in Norway in 1992 and 1996. *J Antimicrob Chemother* **43:** 243–252.

Grave, K., Jensen, V.F., McEwen, S., and Kruse, H. (2006) Monitoring of antimicrobial drug usage in animals: methods and applications. In *Antimicrobial Resistance in Bacteria of Animal Origin*. Aarestrup, F.M. (ed.). Washington, DC, USA: ASM Press, pp. 375–395.

Groh, J.L., Luo, Q., Ballard, J.D., and Krumholz, L.R. (2007) Genes that enhance the ecological fitness of *Shewanella oneidensis* MR-1 in sediments reveal the value of antibiotic resistance. *Appl Environ Microbiol* **73:** 492–498.

Guardabassi, L., Dalsgaard, A., Raffatellu, M., and Olsen, J.E. (2000) Increase in the prevalence of oxolinic acid resistant *Acinetobacter* spp observed in a stream receiving the effluent from a freshwater trout farm following the treatment with oxolinic acid-medicated feed. *Aquaculture* **188:** 205–218.

Guerin, E., Cambray, G., Sanchez-Alberola, N., Campoy, S., Erill, I., Da Re, S., *et al*. (2009) The SOS response controls integron recombination. *Science* **324:** 1034.

Guglielmetti, E., Korhonen, J.M., Heikkinen, J., Morelli, L., and von Wright, A. (2009) Transfer of plasmid-mediated resistance to tetracycline in pathogenic bacteria from fish and aquaculture environments. *FEMS Microbiol Lett* **293:** 28–34.

Guglielmini, J., Quintais, L., Garcillán-Barcia, M.P., de la Cruz, F., and Rocha, E.P.C. (2011) The repertoire of ICE in prokaryotes underscores the unity, diversity, and ubiquity of conjugation. *PLoS Genet* **7:** e1002222.

Gullberg, E., Cao, S., Berg, O.G., Ilback, C., Sandegren, L., Hughes, D., and Andersson, D.I. (2011) Selection of resistant bacteria at very low antibiotic concentrations. *PLoS Pathog* **7:** e1002158.

Hall, R.M. (2010) Salmonella genomic islands and antibiotic resistance in *Salmonella enterica*. *Fut Microbiol* **5:** 1525–1538.

Hansen, P.K., Lunestad, B.T., and Samuelsen, O.B. (1993) Effects of oxytetracycline, oxolinic acid, and flumequine on

bacteria in an artificial marine fish farm sediment. *Can J Microbiol* **39:** 1307–1312.
Hastein, T., Hjeltnes, B., Lillehaug, A., Utne Skare, J., Berntssen, M., and Lundebye, A.K. (2006) Food safety hazards that occur during the production stage: challenges for fish farming and the fishing industry. *Rev Sci Tech* **25:** 607–625.
Hastings, P.J., Rosenberg, S.M., and Slack, A. (2004) Antibiotic-induced lateral transfer of antibiotic resistance. *Trends Microbiol* **12:** 401–404.
Haya, K., Burridge, L.E., and Chang, B.D. (2001) Environment impact of chemical wastes produced by the salmon aquaculture industry. *ICES J Mar Sci* **58:** 492–496.
Hazen, T.H., Pan, L., Gu, J.D., and Sobecky, P.A. (2010) The contribution of mobile genetic elements to the evolution and ecology of Vibrios. *FEMS Microbiol Ecol* **74:** 485–499.
Heepngoen, P., Sajjaphan, K., Ferguson, J.A., and Sadowsky, M.J. (2008) Genetic and physiological characterization of oxytetracycline-resistant bacteria from giant prawn farms. *J Microbiol Biotechnol* **18:** 199–206.
Hehemann, J.H., Correc, G., Barbeyron, T., Helbert, W., Czjzek, M., and Michel, G. (2010) Transfer of carbohydrate-active enzymes from marine bacteria to Japanese gut microbiota. *Nature* **464:** 908–912.
Hehemann, J.H., Kelly, A.G., Pudlo, N.A., Martens, E.C., and Boraston, A.B. (2012) Bacteria of the human gut microbiome catabolize red seaweed glycans with carbohydrate-active enzyme updates from extrinsic microbes. *Proc Natl Acad Sci USA* **109:** 19786–19791.
Hektoen, H., Berge, J.A., Hormazabal, V., and Yndestad, M. (1995) Persistence of antibacterial agents in marine sediments. *Aquaculture* **133:** 175–184.
Henríquez-Núnez, H., Evrard, O., Kronvall, G., and Avendaño-Herrera, R. (2012) Antimicrobial susceptibility and plasmid profiles of *Flavobacterium psychrophilum* strains isolated in Chile. *Aquaculture* **354–355:** 38–44.
Hernández, A., Sánchez, M.B., and Martínez, J.L. (2011) Quinolone resistance: much more than predicted. *Front Microbiol* **2:** 1–6.
Herwig, R.P., and Gray, J.P. (1997) Microbial response to antibacterial treatment in marine microcosms. *Aquaculture* **152:** 139–154.
Herwig, R.P., Gray, J.P., and Weston, D.P. (1997) Antibacterial resistant bacteria in superficial sediments near salmon net-cage farms in Puget Sound, Washington. *Aquaculture* **149:** 163–283.
Heuer, O.E., Kruse, H., Grave, K., Collignon, P., Karunasagar, I., and Angulo, F.J. (2009) Human health consequences of use of antimicrobial agents in aquaculture. *Clin Infect Dis* **49:** 1248–1253.
Hill, K.E., Weightman, A.J., and Fry, J.C. (1992) Isolation and screening of plasmids from the epilithon which mobilize recombinant plasmid pD10. *Appl Environ Microbiol* **58:** 1292–1300.
Hoa, P.T.P., Managaki, S., Nakada, N., Takada, H., Anh, D.H., Viet, P.H., and Suzuki, S. (2008) Occurrence rates of sulfamethoxazole and erythromycin-resistant bacteria and drug concentrations in wastewater of integrated aquaculture-agriculture (VAC) sites in Northern Vietnam. In *Interdisciplinary Studies on Environmental Chemistry-Biological Responses to Chemical Pollutants*. Murakami, Y., Nakayama, K., Kitamura, S.I., Iwata, H., and Tanabe, S. (eds). Tokyo, Japan: Terrapub, pp. 355–359.
Hocquet, D., Llanes, C., Thouverez, M., Kulasekara, H.D., Bertrand, X., Plesiat, P., *et al*. (2012) Evidence for induction of integron-based antibiotic resistance by the SOS response in a clinical setting. *PLoS Pathog* **8:** e1002778.
Holmström, K., Gräslund, S., Wahlström, A., Poungshompoo, S., Bengstsson, B.-E., and Kautsky, N. (2003) Antibiotic use in shrimp farming and implications for environment impacts and human health. *Int J Food Sci Technol* **38:** 255–266.
Holten Lützhøft, H.-C., Halling-Sørensen, B., and Jørgensen, S.E. (1999) Algal toxicity of antibacterial agents applied in Danish fish farming. *Arch Environ Contam Toxicol* **436:** 1171–1175.
Hughes, V.M., and Datta, N. (1983) Conjugative plasmids in bacteria of the 'pre-antibiotic' era. *Nature* **302:** 725–726.
Humeniuk, C., Arlet, G., Gautier, V., Grimont, P., Labia, R., and Philippon, A. (2002) Beta-lactamases of *Kluyvera ascorbata*, probable progenitors of some plasmid-encoded CTX-M types. *Antimicrob Agents Chemother* **46:** 3045–3049.
Husevåg, B., Lunestad, B.T., Johnnessen, P.J., Enger, Ø., and Samuelsen, O.B. (1991) Simultaneous occurrence of *Vibrio salmonicida* and antibiotic-resistant bacteria in sediments at abandoned aquaculture sites. *J Fish Dis* **14:** 631–640.
Ibieta, P., Tapia, V., Venegas, C., Hausdorf, M., and Takle, H. (2011) Chilean salmon farming on the horizon of sustainability: review of the development of a highly intensive production, the ISA crisis and implemented actions to reconstruct a more sustainable aquaculture industry. In *Aquaculture and the Environment – A Shared Destiny*. Sladonja, B. (ed.). Rijeka, Croatia: InTech, pp. 215–246.
Ishida, Y., Ahmed, A.M., Mahfouz, N.B., Kimura, T., El-Khodery, S.A., Moawad, A.A., and Shimamoto, T. (2010) Molecular analysis of antimicrobial resistance in Gram-negative bacteria isolated from fish farms in Egypt. *J Vet Med Sci* **76:** 727–734.
Iwamoto, M., Ayers, T., Mahon, B.E., and Swerdlow, D.L. (2010) Epidemiology of seafood-associated infections in the United States. *Clin Microbiol Rev* **23:** 399–411.
Izumi, S., and Aranishi, F. (2004) Relationship between *gyrA* mutations and quinolone resistance in *Flavobacterium psychrophilum* isolates. *Appl Environ Microbiol* **70:** 3968–3972.
Jacobs, L., and Chenia, H.Y. (2007) Characterization of integrons and tetracycline resistance determinants in *Aeromonas* spp. isolated from South African aquaculture systems. *Int J Food Microbiol* **114:** 295–306.
Juhas, M., van der Meer, J.R., Gaillard, M., Harding, R.M., Hood, D.W., and Crook, D.W. (2009) Genomic islands: tools of bacterial horizontal gene transfer and evolution. *FEMS Microbiol Rev* **33:** 376–393.
Kämpfer, P., Lodders, N., Martin, K., and Avendaño-Herrera, R. (2012) *Flavobacterium chilense* sp. nov. and *Flavobacterium araucananum* sp. nov., isolated from farmed salmonid fish. *Int J Syst Evol Microbiol* **62:** 1402–1408.
Kapetanaki, M., Kerry, J., Hiney, M., O'Brien, C., Coyne, R., and Smith, P. (1995) Emergence, in oxytetracycline-free

marine mesocosms, of microorganisms capable of colony formation on oxytetracycline-containing media. *Aquaculture* **134:** 227–236.

Kautsky, N., Rönnbäck, P., Tedengren, M., and Troell, M. (2000) Ecosystem perspectives on management of disease in shrimp pond farming. *Aquaculture* **191:** 145–161.

Kerry, J., Hiney, M., Coyne, R., NicGabhainn, S., Gilroy, D., Cazabon, D., and Smith, P. (1995) Fish feed as a source of oxytetracycline-resistant bacteria in the sediments under fish farms. *Aquaculture* **131:** 101–113.

Kerry, J., Coyne, R., Gilroy, D., Hiney, M., and Smith, P. (1996) Spatial distribution of oxytetracycline and elevated frequencies of oxytetracycline resistance in sediments beneath a marine salmon farm following oxytetracycline therapy. *Aquaculture* **145:** 31–39.

Kibenge, F.S.B., Godoy, M.G., Wang, Y., Kibenge, M.J., Gherardelli, V., Mansilla, S., *et al*. (2009) Infectious salmon anaemia virus (ISAV) isolated from the ISA disease outbreaks in Chile diverged from ISAV isolates from Norway around 1996 and was disseminated around 2005, based on surface glycoprotein gene sequences. *Virol J* **6:** 88.

Kim, E.H., and Aoki, T. (1996a) Sequence analysis of the florfenicol resistance gene encoded in the transferable R-plasmid of a fish pathogen, *Pasteurella piscicida*. *Microbiol Immunol* **40:** 665–669.

Kim, E.H., and Aoki, T. (1996b) Sulfonamide resistance gene in a transferable R plasmid of *Pasteurella piscicida*. *Microbiol Immunol* **40:** 397–399.

Kim, J.H., Hwang, S.Y., Son, J.S., Han, J.E., Jun, J.W., Shin, S.P., *et al*. (2011) Molecular characterization of tetracycline- and quinolone-resistant *Aeromonas salmonicida* isolated in Korea. *J Vet Sci* **12:** 41–48.

Kim, S.K., Nonaka, L., and Suzuki, S. (2004) Occurrence of tetracycline resistance genes *tet(M)* and *tet(S)* in bacteria from marine aquaculture sites. *FEMS Microbiol Lett* **237:** 147–156.

Knapp, C.W., Engemann, C.A., Hanson, M.L., Keen, P.L., Hall, K.J., and Graham, D.W. (2008) Indirect evidence of transposon-mediated selection of antibiotic resistance genes in aquatic systems at low-level oxytetracycline exposures. *Environ Sci Technol* **42:** 5348–5353.

Koenig, J.E., Boucher, Y., Charlebois, R.L., Nesbo, C., Zhaxybayeva, O., Bapteste, E., *et al*. (2008) Integron-associated gene cassettes in Halifax Harbour: assessment of a mobile gene pool in marine sediments. *Environ Microbiol* **10:** 1024–1038.

Koeypudsa, W., Yakupitiyage, A., and Tangtrongpiros, J. (2005) The fate of chlortetracycline residues in a simulated chicken-fish integrated farming systems. *Aquac Res* **36:** 570–577.

Kohanski, M.A., Dwyer, D.J., Hayete, B., Lawrence, C.A., and Collins, J.J. (2007) A common mechanism of cellular death induced by bactericidal antibiotics. *Cell* **130:** 797–810.

Kohanski, M.A., DePristo, M.A., and Collins, J.J. (2010) Sublethal antibiotic treatment leads to multidrug resistance via radical-induced mutagenesis. *Mol Cell* **37:** 311–320.

Kristiansson, E., Fick, J., Janzon, A., Grabic, R., Rutgersson, C., Weijdegard, B., *et al*. (2011) Pyrosequencing of antibiotic-contaminated river sediments reveals high levels of resistance and gene transfer elements. *PLoS ONE* **6:** e17038.

Kruse, H., and Sørum, H. (1994) Transfer of multiple drug resistance plasmids between bacteria of diverse origins in natural microenvironments. *Appl Environ Microbiol* **60:** 4015–4021.

Kummerer, K. (2009) Antibiotics in the aquatic environment – a review – part I. *Chemosphere* **75:** 417–434.

L'Abee-Lund, T.M., and Sørum, H. (2001) Class 1 integrons mediate antibiotic resistance in the fish pathogen *Aeromonas salmonicida* worldwide. *Microb Drug Resist* **7:** 263–272.

L'Abee-Lund, T.M., and Sørum, H. (2002) A global non-conjugative TetC plasmid, pRAS3, from *Aeromonas salmonicida*. *Plasmid* **47:** 172–181.

Lai, H.T., and Lin, J.J. (2009) Degradation of oxolinic acid and flumequine in aquaculture pond waters and sediments. *Chemosphere* **75:** 462–468.

Lang, A.S., Zhaxybayeva, O., and Beatty, J.T. (2012) Gene transfer agents: phage-like elements of genetic exchange. *Nat Rev Microbiol* **10:** 472–482.

Laroche, E., Pawlak, B., Berthe, T., Skurnik, D., and Petit, F. (2009) Occurrence of antibiotic resistance and class 1, 2 and 3 integrons in *Escherichia coli* isolated from a densely populated estuary (Seine, France). *FEMS Microbiol Ecol* **68:** 118–130.

Lau, S.K., Wong, G.K., Li, M.W., Woo, P.C., and Yuen, K.Y. (2008) Distribution and molecular characterization of tetracycline resistance in *Laribacter hongkongensis*. *J Antimicrob Chemother* **61:** 488–497.

Lawrence, J.G. (2000) Clustering of antibiotic resistance genes: beyond the selfish operon. *ASM News* **66:** 281–286.

Le, T.X., and Munekage, Y. (2004) Residues of selected antibiotics in water and mud from shrimp ponds in mangrove areas in Viet Nam. *Mar Pollut Bull* **49:** 922–929.

Le, T.X., Munekage, Y., and Kato, S. (2005) Antibiotic resistance in bacteria from shrimp farming in mangrove areas. *Sci Total Environ* **349:** 95–105.

Le Hello, S., Hendriksen, R.S., Boublet, B., Fisher, I., Møller Nielsen, E., Whichard, J.M., *et al*. (2011) International spread of an epidemic population of *Salmonella enterica* serotype Kentucky ST198 resistant to ciprofloxacin. *J Infect Dis* **204:** 675–684.

León-Muñoz, J., Tecklin, D., Farías, A., and Díaz, S. (2007) *Salmonicultura en los Lagos del Sur de Chile – Ecorregión Valdiviana. Historia, tendencias e impactos medioambientales. [Salmon farming in the Lakes of Southern Chile – Valdivian Ecoregion. History, tendencies and environmental impacts]*. Valdivia, Chile: WWF Chile – Núcleo Científico Milenio Forecos, Universidad Austral de Chile.

Leung, K.Y., Siame, B.A., Tenkink, B.J., Noort, R.J., and Mok, Y.K. (2012) *Edwardsiella tarda* – virulence mechanisms of an emerging gastroenteritis pathogen. *Microbes Infect* **14:** 26–34.

Levesque, C., Piche, L., Larose, C., and Roy, P.H. (1995) PCR mapping of integrons reveals several novel combinations of resistance genes. *Antimicrob Agents Chemother* **39:** 185–191.

Levy, S.B., and Marshall, B. (2004) Antibacterial resistance

worldwide: causes, challenges and responses. *Nat Med* **10:** S122–S129.
Levy, S.B., and Marshall, B. (2010) Antibiotics in food animal production: a forty year debate. *APUA Newsletter* **28:** 1–19.
Lin, C.K. (1989) Prawn culture in Taiwan: what went wrong. *World Aquacult* **20:** 19–20.
Linares, J.F., Gustafsson, I., Baquero, F., and Martinez, J.L. (2006) Antibiotics as intermicrobial signaling agents instead of weapons. *Proc Natl Acad Sci USA* **103:** 19484–19489.
Looft, T., Johnson, T.A., Allen, H.K., Bayles, D.O., Alt, D.P., Stedtfeld, R.D., *et al*. (2012) In-feed antibiotic effects on the swine intestinal microbiome. *Proc Natl Acad Sci USA* **109:** 1691–1696.
Looney, W.J., Narita, M., and Mühlemann, K. (2009) *Stenotrophomonas maltophilia*: an emerging opportunist human pathogen. *Lancet Infect Dis* **9:** 312–323.
López, E., Elez, M., Matic, I., and Blázquez, J. (2007) Antibiotic-mediated recombination: ciprofloxacin stimulates SOS-independent recombination of divergent sequences in *Escherichia coli*. *Mol Microbiol* **64:** 83–93.
Love, D.C., Rodman, S., Neff, R.A., and Nachman, K.E. (2011) Veterinary drug residues in seafood inspected by the European Union, United States, Canada, and Japan from 2000 to 2009. *Environ Sci Technol* **45:** 7232–7240.
Lowry, T., and Smith, S.A. (2007) Aquatic zoonoses associated with food, bait, ornamental, and tropical fish. *J Am Vet Med Assoc* **231:** 876–880.
Luna, G.M., Vignaroli, C., Rinaldi, C., Pusceddu, A., Nicoletti, L., Gabellini, M., *et al*. (2010) Extraintestinal *Escherichia coli* carrying virulence genes in coastal marine sediments. *Appl Environ Microbiol* **76:** 5659–5668.
Lunestad, B.T., and Goksøyr, J. (2010) Reduction in the antibacterial effect of oxytetracycline in sea water by complex formation with magnesium and calcium. *Dis Aquat Org* **9:** 67–72.
Lupo, A., Coyne, S., and Berendonk, T.U. (2012) Origin and evolution of antibiotic resistance: the common mechanisms of emergence and spread in water bodies. *Front Microbiol* **3:** 18.
Ma, D., Hu, Y., Wang, J., Ye, S., and Li, A. (2006) Effects of antibacterials use in aquaculture on biogeochemical processes in marine sediment. *Sci Total Environ* **367:** 273–277.
Ma, Y., Paulsen, I.T., and Palenik, B. (2012) Analysis of two marine metagenomes reveals the diversity of plasmids in oceanic environments. *Environ Microbiol* **14:** 453–566.
McDaniel, L.D., Young, E., Delaney, J., Ruhnau, F., Ritchie, K.B., and Paul, J.H. (2010) High frequency of horizontal gene transfer in the oceans. *Science* **330:** 50.
McIntosh, D., Cunningham, M., Ji, B., Fekete, F.A., Parry, E.M., Clark, S.E., *et al*. (2008) Transferable, multiple antibiotic and mercury resistance in Atlantic Canadian isolates of *Aeromonas salmonicida* subsp. *salmonicida* is associated with carriage of an IncA/C plasmid similar to the *Salmonella enterica* plasmid pSN254. *J Antimicrob Chemother* **61:** 1221–1228.
Markestad, A., and Grave, K. (1997) Reduction of antibacterial drug use in Norwegian fish farming due to vaccination. *Dev Biol Stand* **90:** 365–369.
Marshall, B.M., and Levy, S.B. (2011) Food animals and antimicrobials: impacts on human health. *Clin Microbiol Rev* **24:** 718–733.
Marshall, B.M., Ochieng, D.J., and Levy, S.B. (2009) Commensals: underappreciated reservoir of antibiotic resistance. *Microbe* **5:** 231–238.
Marshall, S.H., Henríquez, V., Gómez, F.A., and Cárdenas, C. (2011) ISPsa2, the first mobile genetic element to be described and characterized in the bacterial facultative intracellular pathogen *Piscirickettsia salmonis*. *FEMS Microbiol Lett* **314:** 18–24.
Martinez, J.L. (2009a) The role of natural environments in the evolution of resistance traits in pathogenic bacteria. *Proc Biol Sci* **276:** 2521–2530.
Martinez, J.L. (2009b) Environmental pollution by antibiotics and by antibiotic resistance determinants. *Environ Pollut* **157:** 2893–2902.
Martinez, J.L. (2012) Natural antibiotic resistance and contamination by antibiotic resistance determinants: the two ages in the evolution of resistance to antimicrobials. *Front Microbiol* **3:** 1–3.
Martinez, J.L., Fajardo, A., Garmendia, L., Hernandez, A., Linares, J.F., Martínez-Solano, L., and Sánchez, M.B. (2009) A global view of antibiotic resistance. *FEMS Microbiol Rev* **33:** 44–65.
Martínez-Martínez, L., Eliecer Cano, M., Manuel Rodríguez-Martínez, J., Calvo, J., and Pascual, A. (2008) Plasmid-mediated quinolone resistance. *Expert Rev Anti Infect Ther* **6:** 685–711.
Mazel, D. (2006) Integrons: agents of bacterial evolution. *Nat Rev Microbiol* **4:** 608–620.
Medini, D., Donati, C., Tettelin, H., Masignani, V., and Rappuoli, R. (2005) The microbial pan-genome. *Curr Opin Genet Dev* **15:** 589–594.
Meibom, K.L., Blokesch, M., Dolganov, N.A., Wu, C.Y., and Schoolnik, G.K. (2005) Chitin induces natural competence in *Vibrio cholerae*. *Science* **310:** 1824–1827.
Mellon, M., Benbrook, C., and Benbrook, K.L. (2001) *Hogging It. Estimates of Antimicrobial Abuse in Livestock*. Cambridge, MA, USA: Union of Concerned Scientists.
Meunier, D., Boyd, D., Mulvey, M.R., Baucheron, S., Mammina, C., Nastasi, A., *et al*. (2002) *Salmonella enterica* serotype Typhimurium DT 104 antibiotic resistance genomic island I in serotype Paratyphi B. *Emerg Infect Dis* **8:** 430–433.
Michon, A., Allou, N., Chau, F., Podglajen, I., Fantin, B., and Cambau, E. (2011) Plasmidic *qnrA3* enhances *Escherichia coli* fitness in absence of antibiotic exposure. *PLoS ONE* **6:** e24552.
Midtlyng, P.J., Grave, K., and Horsberg, T.E. (2011) What has been done to minimize the use of antibacterial and antiparasitic drugs in Norwegian aquaculture? *Aquac Res* **42:** 28–41.
Millanao, A. (2002) *Estudio cualitativo y cuantitativo de las quinolonas y fluoroquinolonas importadas y autorizadas para uso y disposición en medicina y en veterinaria en Chile, en el período 1998-2001. Consideraciones sobre su impacto para la salud pública y el medio ambiente. [Qualitative and Quantitative Study of Quinolones and Fluorquinolones Imported and Authorized for Medical and*

Veterinary Use and Disposition in Chile, 1998-2001. Considerations of Its Impact on Public Health and the Envrionment]. Valdivia, Chile: Universidad Austral de Chile. [WWW document]. URL http://cybertesis.uach.cl/tesis/uach/2002/fcm645e/doc/fcm645e.pdf.

Millanao, A.B., Barrientos, H.M., Gomez, C.C., Tomova, A., Buschmann, A., Dölz, H., and Cabello, F.C. (2011) Uso inadecuado y excesivo de antibióticos: salud pública y salmonicultura en Chile. [Injudicious and excessive use of antibiotics: public health and salmon aquaculture in Chile]. *Rev Med Chil* **139:** 107–118.

Miller, C., Thomsen, L.E., Gaggero, C., Mosseri, R., Ingmer, H., and Cohen, S.N. (2004) SOS response induction by beta-lactams and bacterial defense against antibiotic lethality. *Science* **305:** 1629–1631.

Miranda, C.D. (2012) Antimicrobial resistance associated with salmonid farming. In *Antimicrobial Resistance in the Environment*. Keen, P.L., and Montforts, M.H.M.M. (eds). Hoboken, NJ, USA: John Wiley & Sons, pp. 423–451.

Miranda, C.D., and Rojas, R. (2007) Occurrence of florfenicol resistance in bacteria associated with two Chilean salmon farms with different history of antibacterial usage. *Aquaculture* **266:** 39–46.

Miranda, C.D., and Zemelman, R. (2001) Antibiotic resistant bacteria in fish from the Concepción Bay, Chile. *Mar Pollut Bull* **42:** 1096–1102.

Miranda, C.D., and Zemelman, R. (2002a) Bacterial resistance to oxytetracycline in Chilean salmon farming. *Aquaculture* **293:** 207–218.

Miranda, C.D., and Zemelman, R. (2002b) Antimicrobial multiresistance in bacteria isolated from freshwater Chilean salmon farms. *Sci Total Environ* **293:** 207–218.

Miranda, C.D., Kehrenberg, C., Ulep, C., Schwarz, S., and Roberts, M.C. (2003) Diversity of tetracycline resistance genes in bacteria from Chilean salmon farms. *Antimicrob Agents Chemother* **47:** 883–888.

Molbak, K. (2006) The clinical importance of animal-related resistance. In *Antimicrobial Resistance in Bacteria of Animal Origin*. Aarestrup, F.M. (ed.). Washington, DC, USA: ASM Press, pp. 329–337.

Morii, H. (2004) Cloning and nucleotide sequence analysis of the ampicillin resistance gene on conjugative R plasmid from the fish pathogen *Photobacterium damselae* subsp. *piscicida*. *J Aquat Anim Health* **16:** 197–207.

Murray, A.G., and Peeler, E.J. (2005) A framework for understanding the potential for emerging diseases in aquaculture. *Prev Vet Med* **67:** 223–235.

Nadirah, M., Najiah, M., and Teng, S.Y. (2012) Characterization of *Edwardsiella tarda* isolated from Asian seabass, *Lates calcarifer*. *Int Food Res J* **19:** 1247–1252.

Navarrete, P., Mardones, P., Opazo, R., Espejo, R., and Romero, J. (2008) Oxytetracycline treatment reduces bacterial diversity of intestinal microbiota of Atlantic salmon. *J Aquat Anim Health* **20:** 177–183.

Navarro, N., Leakey, R.J.G., and Black, K.D. (2008) Effect of salmon cage aquaculture on the pelagic environment of temperate coastal waters: seasonal changes in nutrients and microbial community. *Mar Ecol Prog Ser* **361:** 47–58.

Naviner, M., Gordon, L., Giraud, E., Denis, M., Mangion, C., Le Bris, H., and Ganiére, J.-P. (2011) Antimicrobial resistance of *Aeromonas* spp. Isolated from the growth pond to the commercial product in a rainbow trout farm following a flumequine treatment. *Aquaculture* **315:** 236–241.

Nawaz, M., Khan, S.A., Tran, Q., Sung, K., Khan, A.A., Adamu, I., and Steele, R.S. (2012) Isolation and characterization of multidrug-resistant *Klebsiella* spp. isolated from shrimp imported from Thailand. *Int J Food Microbiol* **155:** 179–184.

Nayak, S.K. (2010) Role of gastrointestinal microbiota in fish. *Aquaculture* **41:** 1553–1573.

Naylor, R., and Burke, M. (2005) Aquaculture and ocean resource: raising tigers of the sea. *Annu Rev Environ Resour* **30:** 185–218.

Ndi, O., and Barton, M. (2012) Antibiotic resistance in animals – the Australian perspective. In *Antimicrobial Resistance in the Environment*. Keen, P.L., and Montforts, M.H.M.M. (eds). Hoboken, NJ, USA: John Wiley & Sons, pp. 265–290.

Ng, L.K., Martin, I., Alfa, M., and Mulvey, M. (2001) Multiplex PCR for the detection of tetracycline resistant genes. *Mol Cell Probes* **15:** 209–215.

Nikaido, H. (2009) Multidrug resistance in bacteria. *Annu Rev Biochem* **78:** 119–146.

Nisha, A.R. (2008) Antibiotic residues – a global health hazard. *Vet World* **1:** 375–377.

Nogales, B., Lanfranconi, M.P., Piña-Villalonga, J.M., and Bosch, R. (2011) Anthropogenic perturbations in marine microbial communities. *FEMS Microbiol Rev* **35:** 275–298.

Novotny, L., Dvorska, L., Lorencova, A., Beran, V., and Pavlik, I. (2004) Fish: a potential source of bacterial pathogens for human beings. *Vet Med – Czech* **49:** 343–358.

Oppegaard, H., and Sørum, H. (1994) *gyrA* Mutations in quinolone-resistant isolates of the fish pathogen *Aeromonas salmonicida*. *Antimicrob Agents Chemother* **38:** 2460–2464.

Osorio, C.R., Marrero, J., Wozniak, R.A., Lemos, M.L., Burrus, V., and Waldor, M.K. (2008) Genomic and functional analysis of ICE*Pda*Spa1, a fish-pathogen-derived SXT-related integrating conjugative element that can mobilize a virulence plasmid. *J Bacteriol* **190:** 3353–3361.

Pan, J.C., Ye, R., Wang, H.Q., Xiang, H.Q., Zhang, W., Yu, X.F., *et al*. (2008) *Vibrio cholerae* O139 multiple-drug resistance mediated by *Yersinia pestis* pIP1202-like conjugative plasmids. *Antimicrob Agents Chemother* **52:** 3829–3836.

Parker, J.L., and Shaw, J.G. (2011) *Aeromonas* spp. clinical microbiology and disease. *J Infect* **62:** 109–118.

Pelletier, N., Tyedmers, P., Sonesson, U., Scholz, A., Ziegler, F., Flysjo, A., *et al*. (2009) Not all salmon are created equal: life cycle assessment (LCA) of global salmon farming systems. *Environ Sci Technol* **43:** 8730–8736.

Petersen, A., Andersen, J.S., Kaewmak, T., Somsiri, T., and Dalsgaard, A. (2002) Impact of integrated fish farming on antimicrobial resistance in a pond environment. *Appl Environ Microbiol* **68:** 6036–6042.

Pitkanen, L.K., Tamminen, M., Hynninen, A., Karkman, A., Corander, J., Kotilainen, A., and Virta, M. (2011) Fish farming affects the abundance and diversity of the mercury resistance gene *merA* in marine sediments. *Microbes Environ* **26:** 205–211.

Poirel, L., Rodriguez-Martinez, J.M., Mammeri, H., Liard, A., and Nordmann, P. (2005) Origin of plasmid-mediated quinolone resistance determinant QnrA. *Antimicrob Agents Chemother* **49:** 3523–3525.

Poirel, L., Cattoir, V., and Nordmann, P. (2012) Plasmid-mediated quinolone resistance; interactions between human, animal, and environmental ecologies. *Front Microbiol* **3:** 24.

Pulkkinen, K., Suomalainen, L.R., Read, A.F., Ebert, D., Rintamaki, P., and Valtonen, E.T. (2010) Intensive fish farming and the evolution of pathogen virulence: the case of columnaris disease in Finland. *Proc Biol Sci* **277:** 593–600.

Rhodes, G., Huys, G., Swings, J., McGann, P., Hiney, M., Smith, P., and Pickup, R.W. (2000) Distribution of oxytetracycline resistance plasmids between aeromonads in hospital and aquaculture environments: implication of Tn*1721* in dissemination of the tetracycline resistance determinant Tet A. *Appl Environ Microbiol* **66:** 3883–3890.

Rhodes, G., Parkhill, J., Bird, C., Ambrose, K., Jones, M.C., Huys, G., *et al*. (2004) Complete nucleotide sequence of the conjugative tetracycline resistance plasmid pFBAOT6, a member of a group of IncU plasmids with global ubiquity. *Appl Environ Microbiol* **70:** 7497–7510.

Ribeiro, R.V., Reis, E.M., Reis, C.M., Freitas-Almeida, A.C., and Rodrigues, D.P. (2010) Incidence and antimicrobial resistance of enteropathogens isolated from an integrated aquaculture system. *Lett Appl Microbiol* **51:** 611–618.

Roberts, M.C. (2009) The evolution of antibiotic-resistant microbes in foods and host ecosystems. In *Food-Borne Microbes: Shaping the Host Ecosystem*. Jaykus, L.A., Wang, H.H., and Schlesinger, L.S. (eds). Washington, DC, USA: ASM Press, pp. 213–229.

Roberts, M.C., Schwarz, S., and Aarts, H.J.M. (2012) Erratum: acquired antibiotic resistance genes: an overview. *Front Microbiol* **3:** Article 384.

Roberts, M.D., and Schwarz, S. (2009) Tetracycline and chloramphenicol resistance mechanisms. In *Antimicrobial Drug Resistance*. Mayers, D.L. (ed.). New York, NY, USA: Humana Press, pp. 183–193.

Robicsek, A., Strahilevitz, J., Jacoby, G.A., Macielag, M., Abbanat, D., Park, C.H., *et al*. (2006) Fluoroquinolone-modifying enzyme: a new adaptation of a common aminoglycoside acetyltransferase. *Nat Med* **12:** 83–88.

Rodgers, C.J., and Furones, M.D. (2009) Antimicrobial agents in aquaculture: practice, needs and issues. In *The Use of Veterinary Drugs and Vaccines in Mediterranean Aquaculture*. Rodgers, C.J., and Basurco, B. (eds). Zaragoza, Spain: CIHEAM, pp. 41–59.

Rodriguez, M.M., Power, P., Radice, M., Vay, C., Famiglietti, A., Galleni, M., *et al*. (2004) Chromosome-encoded CTX-M-3 from *Kluyvera ascorbata*: a possible origin of plasmid-borne CTX-M-1-derived cefotaximases. *Antimicrob Agents Chemother* **48:** 4895–4897.

Rodriguez-Blanco, A., Lemos, M.L., and Osorio, C.R. (2012) Integrating conjugative elements as vectors of antibiotic, mercury, and quaternary ammonium compound resistance in marine aquaculture environments. *Antimicrob Agents Chemother* **56:** 2619–2626.

Rosewarne, C.P., Pettigrove, V., Stokes, H.W., and Parsons, Y.M. (2010) Class 1 integrons in benthic bacterial communities: abundance, association with Tn*402*-like transposition modules and evidence for coselection with heavy-metal resistance. *FEMS Microbiol Ecol* **72:** 35–46.

Rosser, S.J., and Young, H.K. (1999) Identification and characterization of class 1 integrons in bacteria from an aquatic environment. *J Antimicrob Chemother* **44:** 11–18.

Rossolini, G.M., D'Andrea, M.M., and Mugnaioli, C. (2008) The spread of CTX-M-type extended-spectrum β-lactamases. *Clin Microbiol Infect* **14** (Suppl. 1): 33–41.

Rowe-Magnus, D.A., and Mazel, D. (1999) Resistance gene capture. *Curr Opin Microbiol* **2:** 483–488.

Saga, T., Kaku, M., Onodera, Y., Yamachika, S., Sato, K., and Takase, H. (2005) *Vibrio parahaemolyticus* chromosomal *qnr* homologue *VPA0095*: demonstration by transformation with a mutated gene of its potential to reduce quinolone susceptibility in *Escherichia coli*. *Antimicrob Agents Chemother* **49:** 2144–2145.

SalmonChile (2008) Producción mundial de salmon y trucha cultivado. [WWW document]. URL http://www.salmonchile.cl/files/T4-Mundial%201997-2007.pdf.

Salyers, A., and Shoemaker, N.B. (2006) Reservoirs of antibiotic resistance genes. *Anim Biotechnol* **17:** 137–146.

Samuelsen, O.B. (1989) Degradation of oxytetracycline in seawater at two different temperatures and light intensities, and the persistence of oxytetracycline in the sediment from a fish farm. *Aquaculture* **83:** 7–16.

Samuelsen, O.B., Torsvik, V., and Ervik, A. (1992a) Long-range changes in oxytetracycline concentration and bacterial resistance toward oxytetracycline in a fish farm sediment after medication. *Sci Total Environ* **114:** 25–36.

Samuelsen, O.B., Lunestad, B.T., Husevåg, B., Hølleland, T., and Ervik, A. (1992b) Residues of oxolinic acid in wild fauna following medication in fish farms. *Dis Aquat Org* **12:** 111–119.

Samuelsen, O.B., Lunestad, B.T., Ervik, A., and Fjelde, S. (1994) Stability of antibacterial agents in an artificial marine aquaculture sediment studied under laboratory conditions. *Aquaculture* **126:** 283–290.

Sandaa, R.-A., Torsvik, V.L., and Goksoyr, J. (1992) Transferable drug resistance in bacteria from fish-farm sediments. *Can J Microbiol* **38:** 1061–1065.

Sandoz, K.M., and Rockey, D.D. (2010) Antibiotic resistance in *Chlamydiae*. *Future Microbiol* **5:** 1427–1442.

Sapkota, A., Sapkota, A.R., Kucharski, M., Burke, J., McKenzie, S., Walker, P., and Lawrence, R. (2008) Aquaculture practices and potential human health risks: current knowledge and future priorities. *Environ Int* **34:** 1215–1226.

Sarmah, A.K., Meyer, M.T., and Boxall, A.B. (2006) A global perspective on the use, sales, exposure pathways, occurrence, fate and effects of veterinary antibiotics (VAs) in the environment. *Chemosphere* **65:** 725–759.

Sarria, J.C., Vidal, A.M., and Kimbrough, R.C., 3rd (2001) Infections caused by *Kluyvera* species in humans. *Clin Infect Dis* **33:** e69–74.

Schmidt, A.S., Bruun, M.S., Dalsgaard, I., Pedersen, K., and Larsen, J.L. (2000) Occurrence of antimicrobial resistance in fish-pathogenic and environmental bacteria associated with four danish rainbow trout farms. *Appl Environ Microbiol* **66:** 4908–4915.

Schmidt, A.S., Bruun, M.S., Dalsgaard, I., and Larsen, J.L. (2001a) Incidence, distribution, and spread of tetracycline resistance determinants and integron-associated antibiotic resistance genes among motile aeromonads from a fish farming environment. *Appl Environ Microbiol* **67:** 5675–5682.

Schmidt, A.S., Bruun, M.S., Larsen, J.L., and Dalsgaard, I. (2001b) Characterization of class 1 integrons associated with R-plasmids in clinical *Aeromonas salmonicida* isolates from various geographical areas. *J Antimicrob Chemother* **47:** 735–743.

Seiler, C., and Berendonk, T.U. (2012) Heavy metal driven co-selection of antibiotic resistance in soil and water bodies impacted by agriculture and aquaculture. *Front Microbiol* **3:** Article 399.

Seyfried, E.E., Newton, R.J., Rubert, K.F., Pedersen, J.A., and McMahon, K.D. (2010) Occurrence of tetracycline resistance genes in aquaculture facilities with varying use of oxytetracycline. *Microb Ecol* **59:** 799–807.

Shah, S.Q.A. (2012) Antimicrobial resistance in fish pathogenic and aquatic environmental bacteria. PhD Dissertation. Oslo, Norway: Norwegian School of Veterinary Science.

Shah, S.Q.A., Karatas, S., Nilsen, H., Steinum, T.M., Colquhoun, D.J., and Sørum, H. (2012a) Characterization and expression of the *gyrA* gene from quinolone resistant *Yersinia ruckeri* strains isolated from Atlantic salmon (*Salmo salar* L.) in Norway. *Aquaculture* **350–353:** 37–41.

Shah, S.Q.A., Nilsen, H., Bottolfsen, K., Colquhoun, D.J., and Sørum, H. (2012b) DNA gyrase and topoisomerase IV mutations in quinolone-resistant *Flavobacterium psychrophilum* isolated from diseased salmonids in Norway. *Microb Drug Resist* **18:** 207–214.

Silbergeld, E.K., Graham, J., and Price, L.B. (2008) Industrial food animal production, antimicrobial resistance, and human health. *Annu Rev Public Health* **29:** 151–169.

Silley, P. (2007) Impact of antimicrobial residues on gut communities: are the new regulations effective? *J Appl Microbiol* **102:** 1220–1226.

Silva, J., Zemelman, R., Mandoca, M.A., Henriquez, M., Merino, C., and Gonzalez, C. (1987) Antibiotic-resistant gram negative bacilli isolated from sea water and shellfish. Possible epidemiological implications. *Rev Latinoam Microbiol* **29:** 165–169.

Skippington, E., and Ragan, M.A. (2011) Lateral genetic transfer and the construction of genetic exchange communities. *FEMS Microbiol Rev* **35:** 707–735.

Smith, P. (1996) Is sediment deposition the dominant fate of oxytetracycline used in marine salmonid farms: a review of available evidence. *Aquaculture* **146:** 157–169.

Smith, P. (2008a) Aquaculture and florfenicol resistance in *Salmonella enterica* Typhimurium DT104. *Emerg Infect Dis* **14:** 1327–1328.

Smith, P. (2008b) Antimicrobial resistance in aquaculture. *Rev Sci Tech* **27:** 243–264.

Smith, P., Hiney, M.P., and Samuelsen, O.B. (1994) Bacterial resistance to antimicrobial agents used in fish farming: a critical evaluation of method and meaning. *Annu Rev Fish Dis* **4:** 273–313.

Sobecky, P.A., and Hazen, T.H. (2009) Horizontal gene transfer and mobile genetic elements in marine systems. *Methods Mol Biol* **532:** 435–453.

Sobecky, P.A., Mincer, T.J., Chang, M.C., and Helinski, D.R. (1997) Plasmids isolated from marine sediment microbial communities contain replication and incompatibility regions unrelated to those of known plasmid groups. *Appl Environ Microbiol* **63:** 888–895.

Sommer, M.O., Church, G.M., and Dantas, G. (2010) The human microbiome harbors a diverse reservoir of antibiotic resistance genes. *Virulence* **1:** 299–303.

Sommer, M.O.A., and Dantas, G. (2011) Antibiotics and the resistant microbiome. *Curr Opin Microbiol* **14:** 556–563.

Sørum, H. (2000) Farming of Atlantic salmon – an experience from Norway. *Acta Vet Scand Suppl* **93:** 129–134, and 149–157.

Sørum, H. (2006) Antimicrobial drug resistance in fish pathogens. In *Antimicrobial Resistance in Bacteria of Animal Origin*. Aarestrup, F.M. (ed.). Washington, DC, USA: ASM Press, pp. 213–238.

Sørum, H. (2008) Antibiotic resistance associated with veterinary drug use in fish farms. In *In Improving Farmed Fish Quality and Safety*. Lie, Ø. (ed.). Cambridge, UK: Woodhead Publishing Limited, pp. 157–182.

Sørum, H., L'Abee-Lund, T.M., Solberg, A., and Wold, A. (2003) Integron-containing IncU R plasmids pRAS1 and pAr-32 from the fish pathogen *Aeromonas salmonicida*. *Antimicrob Agents Chemother* **47:** 1285–1290.

Stalder, T., Barraud, O., Casellas, M., Dagot, C., and Ploy, M.C. (2012) Integron involvement in environmental spread of antibiotic resistance. *Front Microbiol* **3:** 119.

Stepanauskas, R., Glenn, T.C., Jagoe, C.H., Tuckfield, R.C., Lindell, A.H., King, C.J., and McArthur, J.V. (2006) Coselection for microbial resistance to metals and antibiotics in freshwater microcosms. *Environ Microbiol* **8:** 1510–1514.

Stewart, G.J., and Sinigalliano, C.D. (1990) Detection of horizontal gene transfer by natural transformation in native and introduced species of bacteria in marine and synthetic sediments. *Appl Environ Microbiol* **56:** 1818–1824.

Stokes, H.W., and Gillings, M.R. (2011) Gene flow, mobile genetic elements and the recruitment of antibiotic resistance genes into Gram-negative pathogens. *FEMS Microbiol Rev* **35:** 790–819.

Strahilevitz, J., Jacoby, G.A., Hooper, D.C., and Robicsek, A. (2009) Plasmid-mediated quinolone resistance: a multifaceted threat. *Clin Microbiol Rev* **22:** 664–689.

Sun, S., Berg, O.G., Roth, J.R., and Andersson, D.I. (2009) Contribution of gene amplification to evolution of increased antibiotic resistance in *Salmonella typhimurium*. *Genetics* **182:** 1183–1195.

Sundell, K., and Wiklund, T. (2011) Effect of biofilm formation on antimicrobial tolerance of *Flavobacterium psychrophilum*. *J Fish Dis* **34:** 373–383.

Suttle, C.A. (2007) Marine viruses – major players in the global ecosystem. *Nat Rev Microbiol* **5:** 801–812.

Suzuki, S. (2010) Tetracycline resistance gene in Asian aquatic environments. In *Interdisciplinary Studies on Environmental Chemistry – Biological Responses to Contaminants*. Hamamura, N., Suzuki, S., Mendo, S., Barrosao, C.M., Iwata, H., and Tanabe, S. (eds). Tokyo, Japan: TERRAPUB, pp. 1–8.

Tacon, A.G., and Metian, M. (2008) Aquaculture feed and food safety. *Ann N Y Acad Sci* **1140:** 50–59.

Tamminen, M., Karkman, A., Corander, J., Paulin, L., and Virta, M. (2011a) Differences in bacterial community composition in Baltic Sea sediment in response to fish farming. *Aquaculture* **313:** 15–23.

Tamminen, M., Karkman, A., Lohmus, A., Muziasari, W.I., Takasu, H., Wada, S., *et al*. (2011b) Tetracycline resistance genes persist at aquaculture farms in the absence of selection pressure. *Environ Sci Technol* **45:** 386–391.

Taylor, N.G., Verner-Jeffreys, D.W., and Baker-Austin, C. (2011) Aquatic systems: maintaining, mixing and mobilising antimicrobial resistance? *Trends Ecol Evol* **26:** 278–284.

Tello, A., Austin, B., and Telfer, T.C. (2012) Selective pressure of antibiotic pollution on bacteria of importance to public health. *Environ Health Perspect* **120:** 1100–1106.

Thomas, C.M., and Nielsen, K.M. (2005) Mechanisms of, and barriers to, horizontal gene transfer between bacteria. *Nat Rev Microbiol* **3:** 711–721.

Toleman, M.A., and Walsh, T.R. (2010) IS*CR* elements are key players in IncA/C plasmid evolution. *Antimicrob Agents Chemother* **54:** 3534.

Toleman, M.A., and Walsh, T.R. (2011) Combinatorial events of insertion sequences and ICE in Gram-negative bacteria. *FEMS Microbiol Rev* **35:** 912–935.

Toleman, M.A., Bennett, P.M., and Walsh, T.R. (2006) IS*CR* elements: novel gene-capturing systems of the 21st century? *Microbiol Mol Biol Rev* **70:** 296–316.

Toranzo, A.E., Magariños, B., and Romalde, J.L. (2005) A review of the main bacterial fish diseases in mariculture systems. *Aquaculture* **246:** 37–61.

Toth, M., Smith, C., Frase, H., Mobashery, S., and Vakulenko, S. (2010) An antibiotic-resistance enzyme from a deep-sea bacterium. *J Am Chem Soc* **132:** 816–823.

Toussaint, A., and Chandler, M. (2012) Prokaryote genome fluidity: toward a system approach of the mobilome. *Methods Mol Biol* **804:** 57–80.

Valdés, I., Jaureguiberry, B., Romalde, J.L., Toranzo, A.E., Margariños, B., and Avendaño-Herrera, R. (2009) Genetic characterization of *Streptococcus phocae* strains isolated from Atlantic salmon, *Salmo salar* L., in Chile. *J Fish Dis* **32:** 351–358.

Venter, J.C., Remington, K., Heidelberg, J.F., Halpern, A.L., Rusch, D., Eisen, J.A., *et al*. (2004) Environmental genome shotgun sequencing of the Sargasso Sea. *Science* **304:** 66–74.

Wegener, H.C. (2003) Ending the use of antimicrobial growth promoters is making a difference. *ASM News* **69:** 443–448.

Welch, T.J., Fricke, W.F., McDermott, P.F., White, D.G., Rosso, M.L., Rasko, D.A., *et al*. (2007) Multiple antimicrobial resistance in plague: an emerging public health risk. *PLoS ONE* **2:** e309.

Welch, T.J., Evenhuis, J., White, D.G., McDermott, P.F., Harbottle, H., Miller, R.A., *et al*. (2009) IncA/C plasmid-mediated florfenicol resistance in the catfish pathogen *Edwardsiella ictaluri*. *Antimicrob Agents Chemother* **53:** 845–846.

White, D.G., and McDermott, P.F. (2011) Antimicrobial resistance in food-borne pathogens. In *Food-Borne Microbes: Shaping the Host Ecosystem*. Jaykus, L.A., Wang, H.H., and Schlesinger, L.S. (eds). Washington, DC, USA: ASM Press, pp. 231–264.

Wiedenbeck, J., and Cohan, F.M. (2011) Origins of bacterial diversity through horizontal genetic transfer and adaptation to new ecological niches. *FEMS Microbiol Rev* **35:** 957–976.

Wiens, G.D., Rockey, D.D., Wu, Z., Chang, J., Levy, R., Crane, S., *et al*. (2008) Genome sequence of the fish pathogen *Renibacterium salmoninarum* suggests reductive evolution away from an environmental *Arthrobacter ancestor*. *J Bacteriol* **190:** 6970–6982.

Woo, P.T.K., Bruno, D.W., and Lim, L.H.S. (2002) *Diseases and Disorders of Finfish in Cage Culture*. Wallingford, UK: CABI Publishing, CAB International.

Woodford, N., Turton, J.F., and Livermore, D.M. (2011) Multiresistant Gram-negative bacteria: the role of high-risk clones in the dissemination of antibiotic resistance. *FEMS Microbiol Rev* **35:** 736–755.

Wozniak, R.A., and Waldor, M.K. (2010) Integrative and conjugative elements: mosaic mobile genetic elements enabling dynamic lateral gene flow. *Nat Rev Microbiol* **8:** 552–563.

Wozniak, R.A., Fouts, D.E., Spagnoletti, M., Colombo, M.M., Ceccarelli, D., Garriss, G., *et al*. (2009) Comparative ICE genomics: insights into the evolution of the SXT/R391 family of ICEs. *PLoS Genet* **5:** e1000786.

Wright, G.D. (2010) Antibiotic resistance in the environment: a link to the clinic? *Curr Opin Microbiol* **13:** 589–594.

Wright, M.S., Baker-Austin, C., Lindell, A.H., Stepanauskas, R., Stokes, H.W., and McArthur, J.V. (2008) Influence of industrial contamination on mobile genetic elements: class 1 integron abundance and gene cassette structure in aquatic bacterial communities. *ISME J* **2:** 417–428.

Xia, R., Guo, X., Zhang, Y., and Xu, H. (2010) *qnrVC*-like gene located in a novel complex class 1 integron harboring the IS*CR1* element in an *Aeromonas punctata* strain from an aquatic environment in Shandong Province, China. *Antimicrob Agents Chemother* **54:** 3471–3474.

Xu, H., Broersma, K., Miao, V., and Davies, J. (2011a) Class 1 and class 2 integrons in multidrug-resistant gram-negative bacteria isolated from the Salmon River, British Columbia. *Can J Microbiol* **57:** 460–467.

Xu, H., Miao, V., Kwong, W., Xia, R., and Davies, J. (2011b) Identification of a novel fosfomycin resistance gene (*fosA2*) in *Enterobacter cloacae* from the Salmon River, Canada. *Lett Appl Microbiol* **52:** 427–429.

Zhao, J., and Aoki, T. (1992) Nucleotide sequence analysis of the class G tetracycline resistance determinant from *Vibrio anguillarum*. *Microbiol Immunol* **36:** 1051–1060.