# Reduced environmental impact of antibacterial agents applied in fish farms using the LiftUp feed collector system or a hydroacoustic feed detector

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ABSTRACT: High-performance liquid chromatography was used to quantify residues of oxolinic acid and flumequine in muscle of wild fish caught in the vicinity of fish farms using the LiftUp feed collector system or a hydroacoustic feed detector during medication. Both systems are designed to minimise feed waste and thereby the amount of medicated feed entering the surroundings. The result indicates that both systems decrease the supply of medicated pellets to the wild fish during medication, since the mean and maximum concentrations of drugs in muscle of wild fish were reduced compared to fish farms not using this equipment. Therefore, the equipment will reduce the environmental impact of antibacterial agents used in fish farming.

KEY WORDS: Aquaculture · Antibacterial agents · Environment

## **INTRODUCTION**

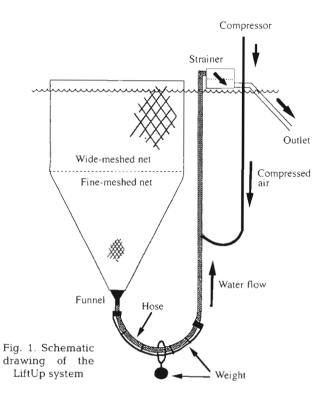
Previous research has shown that during medication in fish farms, drugs reach the wild fauna (Møster 1986, Björklund et al. 1990, 1991, Samuelsen et al. 1992, Ervik et al. 1994). Possible sources are excess food pellets due to overfeeding, small drug-containing feed particles and drug-containing faeces due to incomplete absorption of the drugs in the fish intestine (Cravedi et al. 1987, Gröndel et al. 1987, Hustvedt et al. 1991). Since observations using underwater cameras indicate that wild fish usually do not eat faeces (J. E. Fosseidengen pers. comm.), excess pellets are thought to be the most important source. In order to reduce drug residues in wild fish during medication, pellets available to the wild fish must be minimised. The LiftUp feed collector system and the hydroacoustic feed detector are both designed to minimise feed waste in fish farming (Birkeland & Johnsen 1991, Juell et al. 1993). In this investigation we examined the spread of drugs to wild fish captured in the vicinity of fish farms using either the LiftUp feed collector or the hydroacoustic feed detector during medication.

### MATERIALS AND METHODS

**Chemicals.** Oxolinic acid (OXA) and flumequine (FLU) were obtained from Norsk Medisinaldepot A/S (Bergen, Norway). Methanol, acetonitrile, tetra-hydrofuran (HPLC-grade), dichloromethane, sodium hydroxide and oxalic acid dihydrate (pa-grade) were all from Merck (Darmstadt, Germany).

Sampling sites. Muscle samples of wild fish were collected at 3 marine fish farms located on the west coast of Norway in the Hordaland region during 1992. A feed detector was installed at Farm 1 while the LiftUp system was used in Farms 2 and 3. At Farm 1, 4000 fish with an average weight of 250 g were treated with 160 kg Aqualets containing a total of 5 kg FLU. The medication took place from 2 to 11 October 1992. The seawater temperature was 14 °C on average. At Farm 2, 5880 fish with an average weight of 1.1 kg were given 210 kg pelleted dry feed (7 mm) containing a total of 3.3 kg of OXA. The medication took place from 21 to 30 June 1992. The seawater temperature was 16°C on average. At Farm 3, 370 fish with an average weight of 4.1 kg were given 35 kg pelleted dry feed (9 mm) containing a total of 1.75 kg of OXA. The medication took place from 9 to 18 August 1992. The seawater temperature was 12.5 °C on average.

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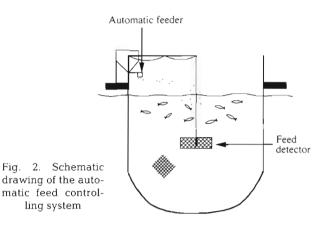


LiftUp. LiftUp is a netting cage consisting of a finemeshed funnel-shaped lower part and a wide-meshed upper part. A schematic drawing of the system is shown in Fig. 1. Dead fish, excess pellets and large faecal particles are trapped by the lower fine-meshed net and led to the bottom of the net where a hose is connected. By introducing a stream of compressed air into the bottom of the hose, the solid material is forced upward to the surface and separated by the strainer. LiftUp is very efficient, collecting nearly 100% of the waste food particles sized 6 mm or larger and nearly 70% of 4 mm particles (Birkeland & Johnsen 1991).

**Feed detector.** A feed detector developed for automatic feeding control in sea-cage rearing of salmon was used (Juell et al. 1993). The feed detector is an acoustic transmitter and receiver mounted inside the

rearing cage in a small net-cage and connected to an automatic feeder. When the echo energy from sinking feed pellets passing the detector has reached a preset level, the automatic feeder is temporarily switched off. A schematic drawing of the system is shown in Fig. 2.

**Sampling.** Samples were collected 2 d prior to treatment (Sampling 1), after 5 d of treatment (Sampling 2) and the first day following termination of medication



(Sampling 3). The fish were caught by gill-nets placed within 30 m of the farm or by angling from the farm site. Samples of fish muscle were taken at the farm site and kept on ice during transport to the laboratory. The samples were stored at -20 °C until analysed.

Analysis. Muscle samples were prepared for highperformance liquid chromatography (HPLC) analysis following the procedure of Samuelsen et al. (1992). To quantify the residue of OXA in muscle, FLU was used as an internal standard and vice versa. To analyse the muscle samples the HPLC equipment and chromatographic assay described by Samuelsen (1990) were used. Samples with a muscle concentration exceeding  $0.01 \ \mu g \ g^{-1}$  were considered positive.

### **RESULTS AND DISCUSSION**

The results from Farms 1, 2 and 3 are shown in Tables 1, 2 & 3 respectively. Prior to medication at Farm 1 we found that 17% of the catch contained residues of FLU (Table 1). This was probably due to fish migrating from another fish farm in the area that had recently been medicated with this drug. That fish migrate between fish farms has previously been shown (Bjordal & Skaar 1992, Bjordal & Johnstone 1993). On

Table 1. Muscle residues of flumequine (FLU) in wild fish caught in the vicinity of Farm 1 prior to, during and after medication

Time of sampling	Species	No. of individuals	% Positive	- Mean muscle conc. (μg g <sup>-1</sup> )	Max. muscle conc. (µg g <sup>-1</sup> )
Prior to medication	Saitheª	30	17	0.01	0.03
During medication	Saithe	26	87	0.92	6.86
After medication	Saithe	30	53	0.19	1.83
*Pollachius v	irens				

Species

Saithe<sup>a</sup>

Cod<sup>b</sup>

Time of

Prior to

medication

sampling

the fifth day of medication, 87 % of the catch contained FLU residues with a mean and maximum concentration of 0.92 and 6.86  $\mu q q^{-1}$ respectively. The unexpectedly high percentage of the catch that contained FLU residues and the high concentrations found are best explained by an incorrect adjustment of the automatic feeder, since no other farm in the area was medicating with FLU at the time. During the first 5 d of treatment, pellets were occasionally thrown out of the cage and were therefore easily available to the wild fish. On the fifth day the automatic feeder was adjusted. By the end of medication the number of positive samples had decreased to 53% with a mean and maximum concentration of 0.19 and 1.83  $\mu$ g g<sup>-1</sup>, respectively. These results show how easily drugs are spread to wild fish and how effective HPLC analysis is in revealing this. The large decrease in both mean and maximum concentrations occurring between samplings 2 and 3 can be best explained by excretion of the drug (Rogstad et al. 1993). The results indicate that, when the feed detector is connected to a correctly adjusted automatic feeder, the system effectively minimises the spread of medicated feed to the surroundings.

Apart from 1 sample containing 1.2  $\mu$ g g<sup>-1</sup>, Table 2 shows that low residues of OXA were found in one-third of the catch at Farm 2 prior to medication. After 5 d of medication 42% of the catch contained OXA residues in muscle and in the majority of these samples the concentration was still low. On the day medication was terminated 83% of the catch contained OXA residues, with a mean and maximum concentration of 0.53 and 4.42  $\mu$ g g<sup>-1</sup>, respectively. Every sampling at Farm 2 included some individuals which were atypical in that they contained much higher concentrations of OXA than

Common dab<sup>c</sup> 0.01 0.01 3 33 Lemon sole<sup>d</sup> 0 0 0 4 TOTAL 29 32 0.09 1.21 53 0.75 During Saithe 15 0.09 medication Pollack<sup>e</sup> 10 0 0 0 Cod 3 33 0.01 0.01 Salmon<sup>f</sup> 3 0 0 0 2 50 0.03 0.06 Common dab TOTAL. 33 42 0.06 0.75 85 0.65 4.42 After Saithe 13 medication Cod 75 0.21 0.57 4 100 Mackerel<sup>g</sup> 1 0.25 0.25 83 TOTAL 18 0.53 4.42

<sup>a</sup>Pollachius virens, <sup>b</sup>Gadus morhua, <sup>c</sup>Limanda limanda, <sup>d</sup>Microstomus kitt, <sup>e</sup>Pollachius pollachius, <sup>f</sup>Salmo salar, <sup>g</sup>Scomber scombrus

Table 3. Muscle residues of oxolinic acid (OXA) in wild fish caught in the vicinity of Farm 3 prior to, during and after medication

Time of sampling	- F	No. of dividuals	% Positive	Mean muscle conc. (µg g <sup>-1</sup> )	Max. muscle conc. (µg g <sup>-1</sup> )
Prior to	Saithe <sup>a</sup>	6	0	0	0
medication	Cod <sup>b</sup>	3	0	0	0
	Common dab	<sup>c</sup> 2	0	0	0
	Lemon sole <sup>d</sup>	1	0	0	0
	Haddock <sup>e</sup>	6	0	0	0
	Rainbow trout	1 9	0	0	0
	Whiting <sup>g</sup>	1	0	0	0
	Salmon <sup>h</sup>	1	0	0	0
	TOTAL	29	0	0	0
During	Saithe	4	50	0.21	0.51
medication	Whiting	4	0	0	0
	Rainbow trout	5	40	0.12	0.59
	TOTAL	13	31	0.11	0.59
After	Saithe	22	28	0.02	0.15
medication	Mackerel <sup>1</sup>	11	64	0.05	0.16
	Salmon	1	0	0	0
	Rainbow trout	7	14	0.01	0.01
	TOTAL	41	34	0.02	0.16

<sup>a</sup>Pollachius virens, <sup>b</sup>Gadus morhua, <sup>c</sup>Limanda limanda, <sup>d</sup>Microstomus kitt, <sup>e</sup>Melanogrammus aeglefinus, <sup>f</sup>Oncorhynchus mykiss, <sup>g</sup>Merlangius merlangus, <sup>h</sup>Salmo salar, <sup>i</sup>Scomber scombrus

Table 2. Muscle residues of oxolinic acid (OXA) in wild fish caught in the vicinity of Farm 2 prior to, during and after medication

No. of

individuals

19

3

%

Positive

42

0

Mean muscle

conc. ( $\mu g \ g^{-1}$ )

0.09

0

Max. muscle

conc. ( $\mu g g^{-1}$ )

1.21

0

the others. A possible explanation for both the atypical samples and the high percentage of positive samples found at Farm 2 is migration of wild fish from another fish farm in the area that also medicated with OXA at the same time.

At Farm 3, no drug residues were found in the wild fish population prior to medication (Table 3). During medication 31% of the samples contained OXA with mean and maximum concentrations of 0.11 and 0.59  $\mu$ g g<sup>-1</sup>, respectively. After medication was terminated 34% of the catch contained OXA, with a mean concentration of 0.02  $\mu$ g g<sup>-1</sup> and a maximum concentration of 0.16  $\mu$ g g<sup>-1</sup>. Farm 3 is located in an area with few other fish farms, and the probability of drug-contaminated wild fish migrating from other fish farms is therefore lower than for Farms 1 and 2.

Ervik et al. (1994) suggest that the calculated mean muscle concentration in the wild fish caught in the vicinity of fish farms after medication is terminated depends on the amount of drug used during treatment. In the investigation of Ervik et al. (1994), Farms 5 and 6 used a total of 1.4 kg OXA and 6 kg FLU, respectively, which is comparable to the amount of drugs used in Farms 1, 2 and 3 in this investigation. On the day medication was terminated at Farms 5 and 6, 77 % of the catch contained drug residues at these farms. This is considerably higher than what was found at Farms 1 and 3 in this investigation. The maximum muscle concentration was 6.62 OXA µg g<sup>-1</sup> at Farm 5 and 15.74 FLU  $\mu$ g g<sup>-1</sup> at Farm 6, and the mean values were calculated to be 1.02 and 0.95  $\mu$ g g<sup>-1</sup>, respectively. In approximately 30% of the samples from Farm 5 and 20% of the samples from Farm 6, the muscle concentration of the drugs exceeded 1  $\mu$ g g<sup>-1</sup>, the corresponding values being 6, 5.5 and 0%, respectively, for Farms 1, 2 and 3. Therefore we conclude that the primary source causing the high concentrations of drugs in the wild fish is the medicated feed. The use of LiftUp or a feed detector during medication minimises this source. The equipment will not, however, prevent transfer of drugs to the wild fish via faeces and small drug-containing feed particles. Whether the low concentrations of drugs in wild fish are due to direct contact with faecal and small feed particles, to eating drug contaminated prey or to a combination of both is impossible to determine without further research.

In conclusion, the use of LiftUp or a feed detector reduces the amount of drugs found in wild fish caught in the vicinity of fish farms during and after medication. Combined with a temporary cessation of fishing in the vicinity of fish farms during and after medication, the use of such equipment will reduce the possibility of drug-containing fish being consumed. Another advan-

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tage of using this equipment is a reduced release of medicated pellets to the sediment which is often present under the cages in fish farms.

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#### LITERATURE CITED

- Birkeland, K., Johnsen, G. H. (1991). Vurdering av LiftUp föroppsamlers effekt på utslipp av antibakterielle midler fra fiskeoppdrett. SFT-prosjekt 112/91. Rådgivende biologer AS, Bergen
- Bjordal, Å., Johnstone, A. D. F. (1993). Local movement of saithe (*Pollachius virens* L.) in the vicinity of fish farm cages. ICES mar. Sci. Symp. 196: 143-146
- Bjordal, Å., Skaar, A. B. (1992). Tagging of saithe (*Pollachius virens* L.) at a Norwegian fish farm: preliminary results on migration. C.M.-ICES/G:35
- Björklund, H., Bondestam, J., Bylund, G. (1990). Residues of oxytetracycline in wild fish and sediments from fish farms. Aquaculture 86: 359–367
- Björklund, H., Råbergh, C. M. I., Bylund, G. (1991). Residues of oxolinic acid and oxytetracycline in fish and sediments from fish farms. Aquaculture 97: 85–96
- Cravedi, J.-P., Choubert, G., Delous, G. (1987). Digestibility of chloramfenicol, oxolinic acid and oxytetracycline in rainbow trout and influence of these antibiotics on lipid digestibility. Aquaculture 60: 133–141
- Ervik, A., Thorsen, B., Eriksen, V., Lunestad, B. T., Samuelsen, O. B. (1994). Impact of administering antibacterial agents on wild fish and blue mussels *Mytilus edulis* in the vicinity of fish farms. Dis. aquat. Org. 18: 45–51
- Gröndel, J. L., Nouws, J. F. M., deJong, M., Schutte, A. R., Driessens, F. (1987). Pharmacokinetics and tissue distribution of oxytetracycline in carp (*Cyprinus carpio* L.) following different routes of administration. J. Fish Dis. 10: 153–163
- Hustvedt, S. O., Salte, R., Kvendseth, O., Vassvik, V. (1991). Bioavailability of oxolinic acid in Atlantic salmon (*Salmo salar* L.) from medicated feed. Aquaculture 97: 305–310
- Juell, J. E., Furevik, D. M., Bjordal, Å. (1993). Demand feeding in salmon farming by hydroacoustic food detection. Aquacult. Eng. 12: 155-167
- Møster, G. (1986). Bruk av antibiotika i fiskeoppdrett. Sogn og Fjordane Distriktshøgskule, Sogndal
- Rogstad, A., Ellingsen, O. F., Syvertsen, C. (1993). Pharmacokinetics and bioavailability of flumequine and oxolinic acid after various routes of administration to Atlantic salmon in seawater. Aquaculture 110: 207–220.
- Samuelsen, O. B. (1990). Simple and rapid determination of flumequine and oxolinic acid in salmon (*Salmo salar*) plasma by high-performance liquid chromatography and fluorescence detection. J. Chromatogr. 530: 452–457
- Samuelsen, O. B., Lunestad, B. T., Husevåg, B., Hølleland, T., Ervik, A. (1992). Residues of oxolinic acid in wild fauna following medication in fish farms. Dis. aquat. Org. 12: 111–119

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