



**PLANAR CHROMATOGRAPHY (HPTLC) AS A TOOL FOR QUALITY CONTROL IN SALMONICULTURE:**

**“Dose Control of antibiotics in Salmon feed.”**

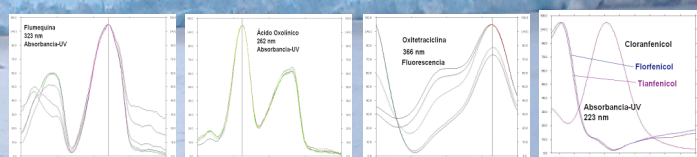
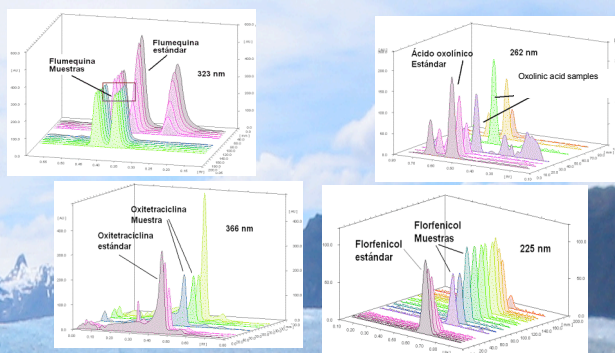
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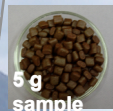
## Introduction

Chile is the second salmon producer in the world exporting over 350,000 Ton/year. Many of destination countries like Japan, USA, and European Community, claim for high quality standards, demanding a salmon feed production safe and controlled. These reasons make necessary to implement analytical methods capable to verify a correct dose of antibacterial and antibiotics incorporated to fish feed.

## Detection



## Extraction



+ 50 mL ACN + KCl/KOH 70 + 30 v/v  
 Nalidixic ac. 0.2 mg/mL. Internal standard

**Flumequine**  
**Oxolinic acid**



+ 50 mL Methanol + HCl (15%)  
 90 + 10, v/v.

**Oxytetracycline**



+ 50 mL Methanol

**Florfenicol**

## Chromatography

Impregnation of HPTLC  
 Sílica gel F<sub>254</sub> plate with

MOBILE PHASE

K<sub>2</sub>HPO<sub>4</sub> 0.1 M  
 in Methanol 50 %

Toluene + Ethylacetate+ Formic acid; 6 + 3 + 1, v/v/v.

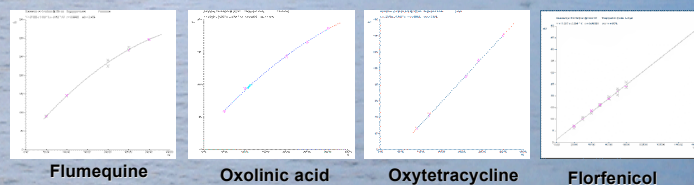
EDTA 5%

Chloroform +MetOH + EDTA 5%  
 65 + 20 + 5, v/v/v.

None

Ethyl acetate + n-Hexane  
 80 + 20, v/v.

## Validation Parameters



Analyte	Linear Range	LOQ	Recovery	Repeatability
Oxytetracycline	50 - 300 ng r <sup>2</sup> : 0.99854	1 ppm	95 %	< 5%
Oxolinic acid	20 - 300 ng r <sup>2</sup> : 0.98995	0.5 ppm	92 %	< 5%
Flumequine	20 - 300 ng r <sup>2</sup> : 0.99712	0.5 ppm	98 %	< 5%
Florfenicol	20 - 200 ng r <sup>2</sup> : 0.99296	5 ppm	90 %	< 5%

Table 1: Illness affecting salmon and antibiotic used for treatment

ILLNESS/ CONDITION	Host	Antibiotics for Treatments
E.R.M. Enteric Red Mouth disease	<i>S. salar</i>	Oxolinic acid, oxytetracycline.
Pseudomonas/ Aeromonas Septicemia	<i>S. salar</i>	Prevention/treatment: Oxytetracycline, Sulphas, Flumequine, Oxolinic acid.
Cold Water Disease	<i>S. salar</i>	Prevention/treatment: Oxytetracycline, Flumequine, Enrofloxacin.
SRS/UA Salmon Rickettsial syndrome	<i>S. salar</i>	Oxytetracycline, Oxolinic acid, Flumequine, (Enrofloxacin, Danafloxacin)
BKD Bacterial Kidney Disease	<i>S. salar</i> <i>S. trutta</i>	Erythromycin, Sulphas, Oxytetracycline.

## Conclusions

- Because medication of salmon with antibiotics it is a common practice in salmon culture. Dose control in fish feed become an essential analysis regarding the quality of the final product.
- HPTLC versatility allows simultaneous analysis of a great number of samples, giving fastness and permitting quantification of two or more substances at the same time.
- These reason make HPTLC a very important analytical tool for fish feed quality control and salmon culture in general..