

# CROSS INJECTION OF SALMON AND TROUT WITH PANCREAS DISEASE VIRUS (PDV) AND SLEEPING DISEASE VIRUS (SDV)

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## Introduction

Pancreas disease virus (PDV) isolated from Atlantic salmon (*Salmo salar* L.) and sleeping disease virus (SDV) isolated from rainbow trout (*Oncorhynchus mykiss*) are both characterised as Alpha viruses. Similar histopathological changes in respective hosts caused by respective viruses have been reported from the field and close serological and genetical relation between the two virus isolates have been documented. To further examine the relation between the two virus isolates both species have been injected with each virus isolate under identical experimental conditions.

## Material and methods

**PDV** were originally isolated in CHSE-214 cells during an outbreak of PD on Atlantic salmon in Ireland (Nelson et al. 1995) and the virus had been grown 10 passages in CHSE cells at 14°C. Injection dose was  $7.8 \log_{10} \text{TCID}_{50} / \text{fish}$ .

**SDV** were originally isolated in CHSE-214 cells during an outbreak of SD on rainbow trout in France (Castric et al. 1997) and the virus had been grown a few passages in RTG-2 cells and 3 passages in CHSE cells at 10°C. Injection dose was  $7.3 \log_{10} \text{TCID}_{50} / \text{fish}$ .

Three groups of 40 Atlantic salmon (20g) mixed with 40 rainbow trout (15g) were injected i.p. with PDV, SDV and saline and held in fresh water at 14°C. Blood samples for antibody analysis were taken pre-injection and 6 weeks post injection (p.i.). Samples of kidney tissue for virus re-isolation were taken 1 week p.i. and samples of pancreas, heart and skeletal muscle tissue for histopathological examination were collected 2, 3 and 6 weeks p.i. A score system was used to evaluate the severity of virus induced lesions (no: -, minimal: +, mild: ++, moderate: +++, severe: +++++). Scores ++ or higher was compatible with virus infection. Statistical analysis of the results was performed by non-parametric statistics (Kruskal Wallis one-way analysis of variance).

## Results

Antibodies with neutralizing activity against PDV were detected 6 w.p.i. in all virus injected groups (Table 1). Neutralizing antibodies against SDV were examined only for trout (because of scarcity of serum) and was detected in fish post injection with both PDV and SDV. The results indicate highly serological cross reactivity between the two virus isolates.

Virus was re-isolated from samples of all virus injected groups 1 w.p.i. (Table 1). More severe CPE (no vacuolisation but heavy necrosis) was seen in CHSE cultures inoculated with samples from SDV injected fish compared to PDV injected fish, indicating different cytopathological courses of the two virus isolates.

No mortality or gross pathology was observed for any group during a period of 6 weeks p.i. Highest level of histopathology score was seen 2 w.p.i. in pancreas and heart tissue and 6 w.p.i. in lateral muscle tissue. By comparing PDV injected and non-injected groups significant differences were seen in all three tissue in both fish species (Table 2). By injection with SDV significant differences were seen in all three tissues in trout but only for pancreas tissue in salmon. By comparing injection with PDV and SDV in each fish species no significant difference with the two viruses was observed in trout while injection with SDV resulted in significant higher score in muscle tissue in trout than in salmon.

Group no	Virus	Fish species	Frequency of viruspositive fish 1 w.p.i.	Frequency of fish with anti- PDV 6 w.p.i.	Frequency of fish with anti- SDV 6 w.p.i.
1	PDV	trout	5/5	7/10	4/5
2	"	salmon	5/5	10/10	NT
3	SDV	trout	5/5	7/10	5/5
4	"	salmon	5/5	5/10	NT
5	no	trout	0/5	0/10	0/5
6	no	salmon	0/5	0/10	NT

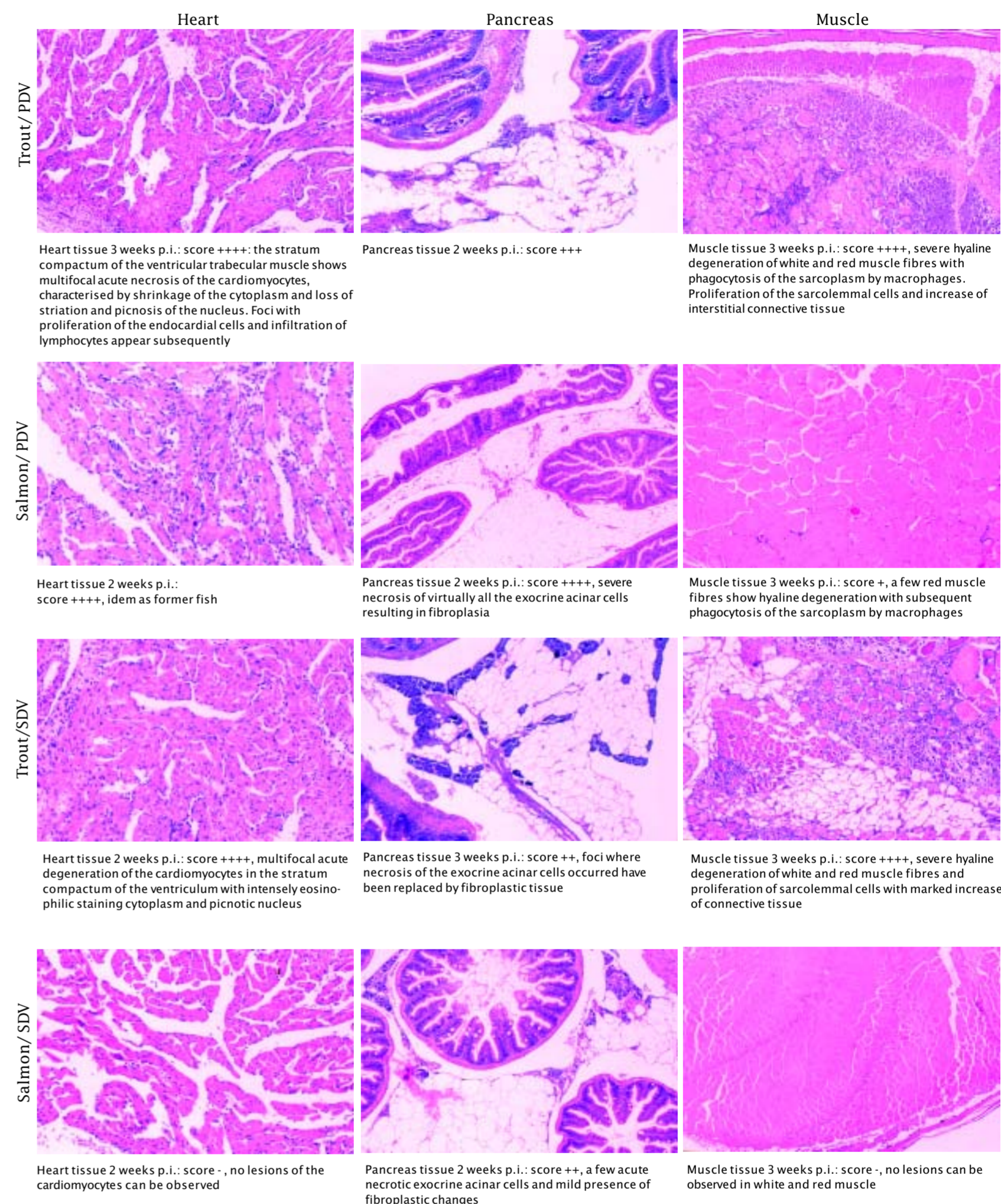
**Table 1.** Virus re-isolation and neutralizing antibodies post injection

Test groups compared	Heart tissue 2 w.p.i.	Pancreas tissue 2 w.p.i.	Muscle tissue 6 w.p.i.
PDV inj. trout/uninj.trout	P<0.05	P<0.01	P<0.01
PDV inj. salmon/uninj.salmon	P<0.01	P<0.01	P<0.05
SDV inj. trout/uninj.trout	P<0.01	P<0.01	P<0.01
SDV inj. salmon/uninj.salmon	n.s	P<0.05	n.s
PDV inj. trout/SDV inj.trout	n.s	n.s	n.s
PDV inj. salmon/SDV inj.salmon	n.s	n.s	P<0.05

**Table 2.** Histopathological score post injection. Statistical analysis by comparing groups

n.s.: no significant difference

NT: not tested



## Conclusion

Experimental infection of Atlantic salmon and rainbow trout with cell culture grown PDV or SDV induced no mortality or gross pathology in the fish. Identical histopathology lesions with both virus isolates were seen in rainbow trout with only minor variation in severity. In Atlantic salmon, PDV induced more severe histopathological lesions than SDV. It is, however, difficult to predict whether this is a general feature of all PDV and SDV isolates and there is also some uncertainty as to how much influence the cell-passage history of these viruses has on the severity of the disease induced in different salmonid species.

Neutralizing antibodies against PDV were detected in both salmon and trout serum post injection with any of the virus isolates. Neutralizing antibodies against SDV were detected in trout serum post injection with any of the virus isolates (salmon serum was not tested against SDV). In general a close serological relation between the two virus isolates is demonstrated and cross-protection against both viruses by vaccination with formulations containing one of the virus isolates is expected.

The results support the suggestion that PDV and SDV are closely related but not identical viruses most possibly belonging to the same serotype.