

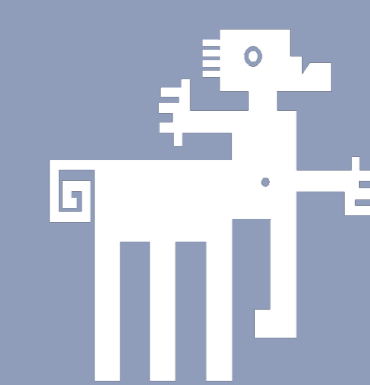
Investigation of the causes of mortality and morbidity in stranded cetaceans on the Catalan coast (June 2021 - May 2022), and a retrospective study of herpesvirus presence in CNS

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1. OBJECTIVES

OBJECTIVE 1

To determine the causes of mortality and morbidity in cetaceans stranded along the Catalan coast between June 2021 and May 2022.

OBJECTIVE 2

To perform a retrospective study on the presence of herpesvirus in the CNS of the cetaceans stranded in the Catalan coast since 2012.

2. MATERIALS AND METHODS



Necropsies (n=2) were performed, as well as further laboratorial studies such as:

- └ Histopathology
- └ Microbiology
- └ IHQ
- └ Biomolecular studies

Frozen samples (n=94) from CNS tissue were analysed for herpesvirus DNA by nested-PCR, following the protocol described by Smolarek Benson et al (2006). The DNA was extracted using the DNeasy Blood & Tissue (Qiagen) and CADOR kits. OvHV-2 was used as a positive control.

3. RESULTS AND DISCUSSION

NECROPSIES

A total of 2 (n=2) necropsies were performed during the period of the study.

Table 1: General data

ID	Species	Sex	Alive or dead; conservation status	Cause of death/stranding	Laboratorial findings
N-334/21	<i>Stenella coeruleoalba</i>	Male	Alive; M2	CeMV infection in CNS; euthanasia.	RT-PCR+ for CeMV in CNS
N-465/21	<i>Stenella coeruleoalba</i>	Female	Dead; M2	By-catch, PUE.	-

D, dead; A, Alive

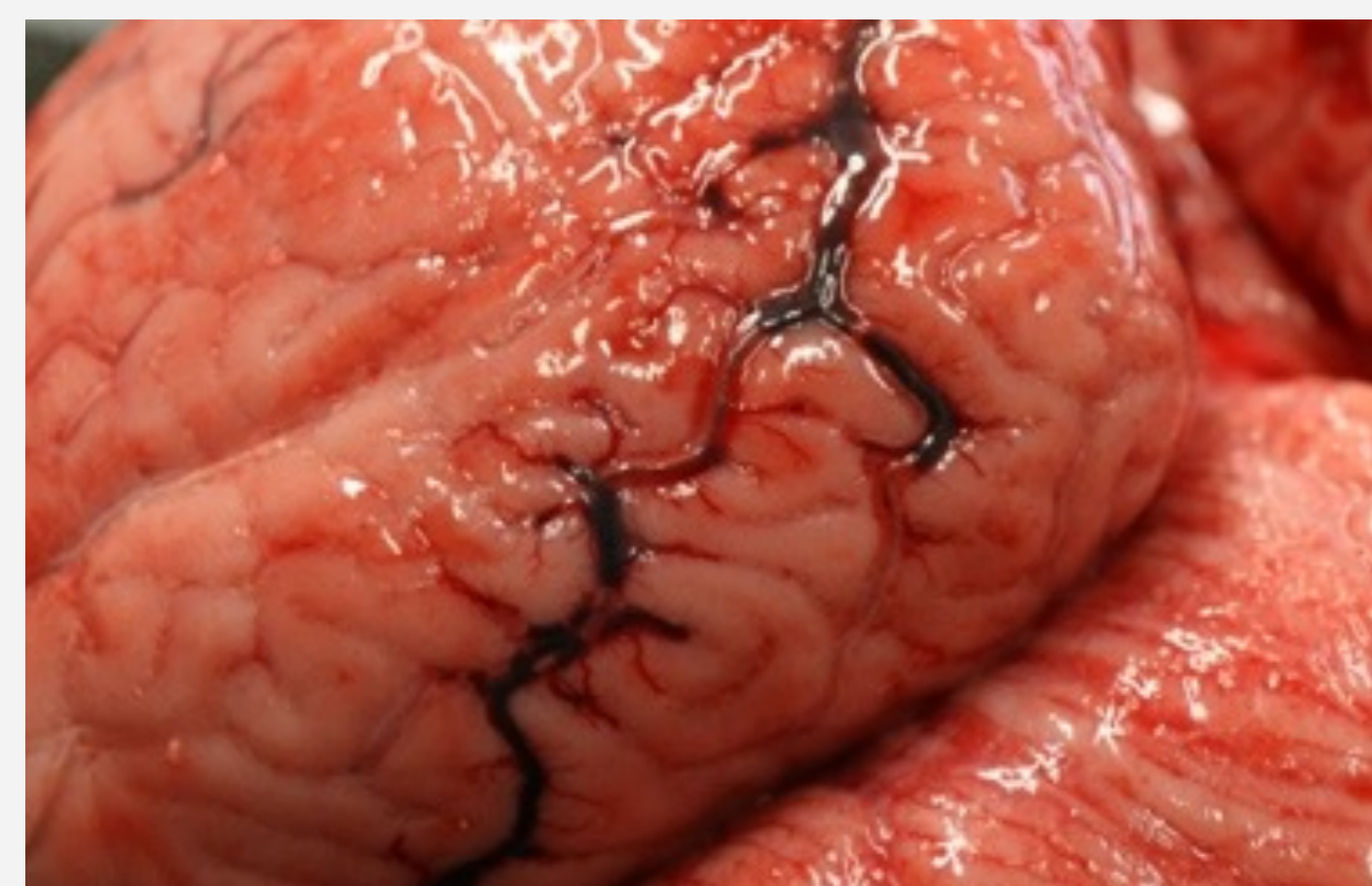


Figure 1: Gas bubbles in the meningeal veins (N-465/21).

N-334/21 - CeMV infection. This striped dolphin presented a mononuclear encephalitis associated with the presence of CeMV antigen, but no other signs of a systemic infection were present. The infection most likely occurred during the 2016-2017 epidemic considering how the lesions were located exclusively in the CNS, characteristic of the chronic manifestation of the disease (Soto et al. 2011; Mira et al. 2019; Cuvertoret-Sanz et al. 2020).

N-465/21 - Human interaction. The lesions present in this other striped dolphin (fishing net marks, disseminated gas bubbles) as well the fact that in the necropsy it was seen that it had recently ingested an abundant amount of food, indicate that the most probable cause of death was by-catch, probably due to fisheries interaction (de Quirós et al. 2018).

NESTED-PCR

A total of 94 frozen CNS samples from 94 (n=94) different individuals were analysed.

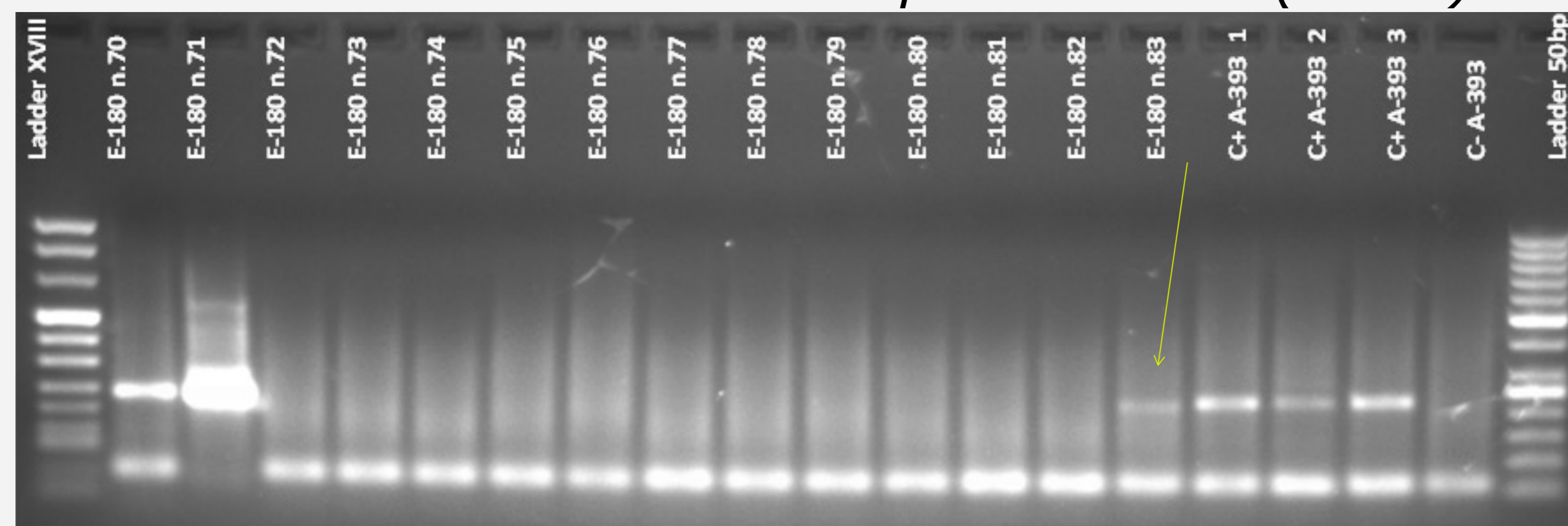


Figure 2: Positive result for herpesvirus infection (green arrow).

The nested-PCR is the most sensitive technique to detect generic herpesvirus (Druce et al. 2002).

The positive result belongs to N-330/12, a striped dolphin (*Stenella coeruleoalba*) whose cause of death was unknown. At the time of the necropsy no lesions suggestive of a herpesvirus infection were detected (Soto et al. 2012). The lack of lesions observed could indicate that the animal was infected but that the infection was latent (Vargas-Castro et al. 2021).

3. CONCLUSIONS

- The causes of cetacean stranding have been CeMV infection and by-catch; both of which have been characterized previously in the Catalan coast as main causes of stranding and death.
- The CeMV case is a chronic infection, there are no signs of a new wave of CeMV circulation.
- Two individuals are not representative of the whole Catalan marine ecosystem.
- The nested-PCR is the most sensitive technique to detect generic herpesviruses.
- The results obtained do not coincide with the ones published in another study performed in the Valencian coast, which could be due to differences when executing the technique as well as to whether the animal had an active or latent infection.

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