

Are squid embryos affected by man-made underwater sound?

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Anthropogenic noise has become a pervasive pollutant in today's oceans and there is now clear evidence that such noise can adversely affect the physiology, morphology, behaviour and survival of marine species. To allow the evaluation of ecosystemic responses to this marine stressor, the type of responses and their magnitudes need to be understood for a wide range of species from different trophic levels. Squids are important components in marine food webs and transfer substantial biomass between pelagic and coastal ecosystems. The effects of noise on squid embryos and potential carry-over effects onto the paralarvae are unknown, to the best of our knowledge, for any cephalopod species. Squids attach their egg strands to physical structures and the embryos are thus unable to move away from stressors during their development. Early life stages are more vulnerable to stressors than the more robust adults of most sessile and mobile species, presenting critical population bottlenecks. It is therefore imperative to consider these stages in marine stressor studies, including noise, if the consequences are to be understood on a population (and ecosystem) level. Our study aims to address the above gap of knowledge, and to generate scientific evidence for informing policy for Scottish/UK and EU environmental legislation.

We tested the hypotheses that chronic exposure of squid embryos (*Loligo* spp.) to ship noise playbacks causes (i) prolonged development time, delayed organogenesis, reduced size of egg strands, egg capsules- and dorsal larval mantle length, (ii) embryonal and larval morphological abnormalities, premature hatching, and reduced survival, (iii) metabolic suppression (D-Glucose, total protein and total phospholipid content of eggs) and (iv) structural damage of sensory epithelia and statolith malformation of freshly hatched larvae. Recently-spawned egg clusters laid onto fishers' ropes (water depth 15-35m) from St Abbs and Eyemouth Voluntary Reserve (North Sea) were transferred to St Abbs Marine Station. Individual egg strands were kept in 8 acoustically transparent 25L plastic bags with constant UV sterilised seawater supply, from a temperature controlled, aerated sump tank. Water parameters in each bag were kept constant ($T=14\pm0.5^{\circ}\text{C}$, salinity 34 ± 0.20 , $\text{DO}>90\%$). Bags in each treatment were held in 1000L water baths and exposed to either 6 hrs of ship noise playback (2 x 3hrs with 2 hrs silence in-between), or to a silent control over 8 hrs, throughout embryonic development (30 days) until the last day of hatching (day 44). Bags were checked hourly, and paralarvae were removed from the experimental bags immediately. Exposure to ship noise playbacks significantly affected squid embryo development, resulting in altered physiology, both premature and delayed hatching, reduced survival, and morphological deformities, compared to the control. The results highlight the importance of considering marine invertebrates when evaluating the effects of anthropogenic noise on marine fauna and their early life history stages. A follow-up field-study is underway.

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