STUNNING OF ARCTIC CHAR (*Salvelinus alpinus*) WITH CARBON DIOXIDE AND ELECTRIC FIELD EXPOSURE: PHYSIOLOGICAL INSIGHTS ON MECHANISMS OF LETHALITY AND WELFARE IMPLICATIONS.

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Carbon dioxide (CO_2) is still widely used to stun fish prior to slaughter in aquaculture, although the method has been criticized for resulting in poor welfare. Alternative methods including electrical stunning, or means of alleviating the negative effects of CO_2, are therefore wanted. Yet, knowledge gaps exist with regards to physiological stress responses and mechanisms of lethality associated with these methods, which constrains the development of alternative techniques. In a series of studies on Arctic char (*Salvelinus alpinus*), an important cold-water aquaculture species in Sweden, cardioventilatory responses and blood physiological indicators of stress were recorded during electrical field exposure, as well as CO_2 exposure alone and in combination with hyperoxia or hypothermia, which may alleviate the adverse effects of CO_2.

A ten minute exposure to CO_2-saturated water at 10°C triggered aversive struggling and escape responses. These were accompanied by gradually reduced heart and ventilation rates, hypotension and moderately increased plasma cortisol levels before equilibrium was irrecoverably lost after ~3 minutes. Cooling to 0.25°C did not markedly affect behavior, cardioventilatory responses or the time until loss of equilibrium during CO_2 exposure, but the increase in plasma cortisol was somewhat exacerbated. Ten minute exposure to 10°C water saturated with combinations of CO_2 and pure oxygen resulted in similar behavioral responses and plasma cortisol increase compared with exposure to pure CO_2. However, the hyperoxic CO_2 exposure resulted in a more variable cardioventilatory response, and all fish subsequently recovered in normoxia, which was not the case with pure CO_2.

Electrical stunning (4V/cm, 125 Hz) for 5 and 30 s resulted in instant immobilization and a ~5 s four-fold blood pressure increase. Cardioventilatory arrest followed the 5 s exposure, but the activity subsequently recovered in ten out of eleven fish. Nevertheless, signs of a systemic stress response were evident after the exposure including hypertension and increased ventilation amplitude and plasma cortisol levels. After the 30 s exposure, cardiac activity initially recovered, but subsequently declined while ventilation never recovered.

Collectively, these data show that neither hypothermia nor additional oxygen can reduce behavioral or physiological stress responses during CO_2 exposure in char, but additional oxygen improved recovery in normoxia. While electrical field exposure rendered the fish instantly immobilized, it appears that circulatory failure due to cardiac ischemia resulting from ventilatory failure is the principal cause of death. Further studies on brain activity (i.e. consciousness) are required to determine the welfare implications of this important finding. Moreover, the dramatic hypertensive spike observed in char may partly explain the hemorrhages that are frequently observed in electrically stunned fish of some species.