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4 1 Modeling the impact of salinity variations on  
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8 2 aquatic environments: including negative and  
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12 3 positive effects in life cycle assessment  
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48 12 **ABSTRACT**  
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52 13 Salinity is changing in aquatic systems due to anthropogenic activities (like irrigation or  
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55 14 dam management) and climate change. Although there are studies on the effects of  
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59 15 salinity variations on individual species, little is known about the effects on overall  
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4 16 ecosystems, these impacts being more uncertain in transitional waters such as estuaries  
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6  
7 17 or fiords. The few works that do address this topic have considered these impacts using  
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10 18 ecotoxicity models. However, these models state that an increase in the concentration of  
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14 19 a pollutant generates an increase in the impacts, disregarding the effects of water  
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17 20 freshening. The present research work introduces a general framework to address the  
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20 21 impacts of salinity variations, including emission-related positive effects. We validated  
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24 22 this framework by applying it to an estuarine area in Galicia (northwestern Spain),  
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27 23 where sharp drops in the salt concentration have caused mass mortalities of shellfish in  
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30 24 recent decades. This research work addresses for the first time the potential effects on  
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34 25 the environment derived from a decrease in the concentration of essential substances,  
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37 26 where the effects of an emission can also generate positive impacts. Moreover, it is  
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40 27 expected that the framework can also be applied to model environmental impacts of  
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44 28 other essential substances in life cycle assessment (LCA), such as metals and  
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47 29 macronutrients.

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51 30 Keywords: Biodiversity, Climate Change, Ecotoxicity, Life Cycle Impact Assessment  
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54 31 (LCIA), Salinity, Species Sensitivity Distribution, Transitional Waters  
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59 32 **SYNOPSIS**  
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4 33 We propose a new method to evaluate the effects of variations, both increases and  
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7 34 decreases, in the concentration of essential substances in aquatic ecosystems.  
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11 35 **INTRODUCTION**  
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15 36 Climate change is altering the biogeochemical cycles on the planet<sup>1</sup> and shifting water  
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18 37 temperature, pH, dissolved oxygen concentration, and salinity<sup>2</sup>. In fact, direct  
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22 38 relationships between anthropogenic CO<sub>2</sub> release and alterations in the water cycle  
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25 39 which result into salinity variations have been already established<sup>3-5</sup>. Other direct  
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29 40 human activities, such as irrigation<sup>6,7</sup>, industrialization and agricultural expansion<sup>8</sup>,  
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32 41 effluent disposal<sup>9,10</sup>, or dam management<sup>11</sup>, are also behind these salinity variations.  
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35 42 These changes and their effects can be even more important in transitional waterbodies,  
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39 43 such as estuaries, deltas, or coastal lagoons, which constitute less than 5% of the  
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42 44 brackish areas worldwide but provide about half of the global fish catch<sup>8</sup>.  
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46 45 However, to the best of our knowledge, just five research works have tried to model the  
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50 46 impacts of salinity changes on the environment<sup>12</sup>. Although a new impact category  
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53 47 addressing salinity was proposed<sup>13-15</sup>, to the best of our knowledge, it has never been  
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56 48 included in an LCA study<sup>16</sup>. Two assessment methodologies focused on soils, modeling  
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59 49 salinity impacts according to variations in soil conductivity and linking them with food  
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4 50 production and crop diversity loss<sup>17,18</sup>. Two other methodologies focused on aquatic  
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7 51 systems, shaping the effects of salinity variations according to ecotoxicity models for  
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10 52 water environments<sup>6,9</sup>. The latter were only partially successful for several reasons, as  
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13 53 ecotoxicity is currently based on the observation that the sensitivities of different  
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16 54 species to a chemical follow a normal distribution, so increased exposure will generate  
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20 55 increased impacts<sup>19</sup>. However, the impact of salinity is not only linked to concentration  
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23 56 increases, but also to concentration decreases (systems can become saltier or fresher), so  
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27 57 an approach based on ecotoxic models would fail to describe these effects. Moreover,  
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30 58 salt is not a pollutant or a toxic, but an essential element, so ecotoxic models might not  
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33 59 be valid to describe its behavior. Hence, a critical improvement of these methodologies  
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37 60 is necessary. Therefore, the aim of this study is to provide a framework for the  
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40 61 evaluation of the impacts of salinity changes in aquatic ecosystems.

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44 62 Estuaries are important ecosystems from an ecological point of view<sup>11</sup>, supporting  
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47 63 highly productive communities of arthropods, mollusks, fish, and birds, as well as  
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51 64 complex food webs<sup>20</sup>. The Western *rías* of the northwest Spanish region of Galicia, vital  
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54 65 for the local economic and social development, are partially mixed estuaries where  
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58 66 partial stratification is maintained by the river discharge in the winter and by solar  
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4 67 heating in the summer<sup>21</sup>. Due to their uniqueness, it is common that geography experts  
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7 68 call these areas by their name in Galician (*rías*)<sup>22</sup>. Nevertheless, in the past few decades,  
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10 69 events of massive shellfish deaths were reported in these estuaries due to sharp  
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13 70 decreases in the concentration of salt in the *rías*<sup>11,23–25</sup>. These occurrences have been  
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17 71 linked to heavy rains, locally managed freshwater releases from river dams, and  
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20 72 increased river runoffs, where the frequency of these climatological episodes is  
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24 73 supposed to increase in the coming years due to climate change<sup>24</sup>. Among the Galician  
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27 74 *rías*, the biggest and most productive one is the *Arousa ría*<sup>11,24,26</sup>. Therefore, this *ría* was  
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30 75 chosen to test and validate the methodology proposed in the present research work.  
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34 76 In sum, we describe in this paper a general model shaping the effects of salt variations  
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38 77 in aquatic systems. Then, we apply the model to a case study for the procedure  
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42 78 validation. By providing this framework, we expect to expand the current knowledge of  
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45 79 transitional water systems, allowing to integrate the evaluation of impacts (positive and  
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48 80 negative) in environmental sustainability assessments, and to improve, for example,  
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51 81 effluent disposal or dam management control in sensitive areas.  
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## 55 82 MATERIALS AND METHODS

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4 83 Salinity is defined as the concentration of several inorganic ions (including Na<sup>+</sup>, Ca<sup>2+</sup>,  
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7 84 Mg<sup>2+</sup>, K<sup>+</sup>, Cl<sup>-</sup>, SO<sub>4</sub><sup>2-</sup>, CO<sub>3</sub><sup>2-</sup>, NO<sub>3</sub><sup>-</sup> and HCO<sub>3</sub><sup>-</sup>)<sup>12,16</sup>, where most of these elements are  
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10 85 essential substances needed for organisms to live. In the present research work, salinity  
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13 86 is referred to as the concentration of NaCl, in kilograms per cubic meter (kg/m<sup>3</sup>), as it is  
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17 87 abundant in water streams considered saline.  
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21 88 Regarding the methodology, impacts linked to chemical releases are measured as in Eq.  
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24 89 (1), where IS is the impact score, CF is the characterization factor, and M the mass of  
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28 90 substance emitted. Then, the CF is calculated considering the principal cause-effect  
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31 91 chains – thus, through fate, exposure, and effect factors (FF, XF and EF, respectively),  
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34 92 according to Eq. (2)<sup>27</sup>.

$$IS = CF \cdot M \quad (1)$$

$$CF = FF \cdot XF \cdot EF \quad (2)$$

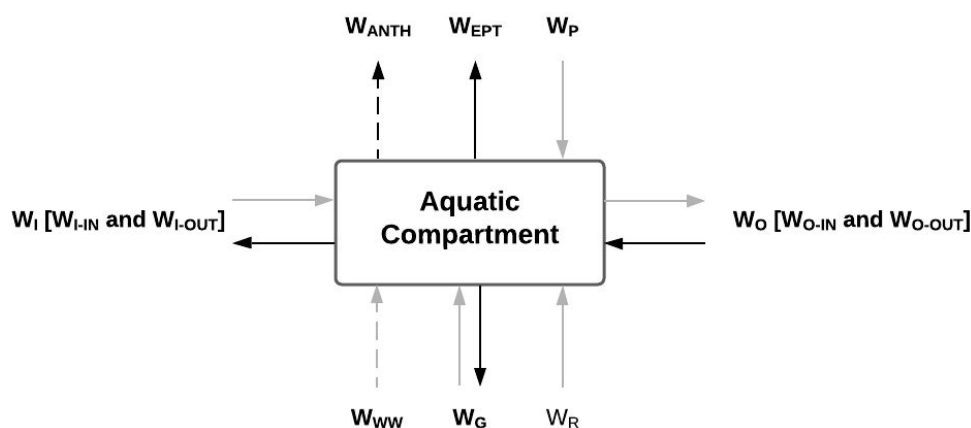
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46 93 CFs addressing impacts on ecosystem quality (aka natural environment) at the endpoint  
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50 94 level have units of potentially disappeared fraction of species (PDF)·m<sup>3</sup>·time/kg. As M  
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53 95 in Eq. (1) is in kilograms, IS has units of PDF·m<sup>3</sup>·time<sup>27</sup>. FF in Eq. (2) is expressed in  
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57 96 units of time (it represents the mass of a chemical in the environment resulting after an  
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60 97 emission flow, so units are kg/(kg/day), which yields days), XF is dimensionless, and

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3 98 EF is expressed as  $\text{PDF} \cdot \text{m}^3/\text{kg}$ . The XF represents the availability of the released  
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7 99 chemical in a system (the fraction of the pollutant that is dissolved in the water), which  
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10 100 can be considered as 1 for salt as it is fully dissolved<sup>9</sup>.

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14 101 - **FATE FACTOR (FF)**

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18 102 The FF is linked to the physical behavior/distribution of the substance in the  
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22 103 environment and normally expresses its persistence in units of time<sup>19,27,28</sup>. However, the  
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26 104 FF also represents the predicted mass residence of a substance in a receiving  
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29 105 compartment per unit of emission flow into it<sup>19</sup>, so the FF is calculated by applying  
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32 106 mass balances to the studied compartment in which degradation and transfer processes  
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36 107 are described<sup>28</sup>. Salt do not degrade (i.e. salt's residence time on the environment equals  
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39 108 millions of years<sup>9</sup>), so the mass balances will describe how they are transferred, and the  
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42 109 relevant flows entering and leaving an aquatic compartment. For the calculation of an  
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46 110 FF for salinity, the streams considered are shown in Figure 1:  
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112 **Figure 1.** Streams to be considered in the mass balances for quantifying the FF. Gray

113 lines represent the inflows and black lines the outflows. Dashed lines are the

114 anthropogenic flows, and continuous lines are natural streams.  $W_i$  refers to each stream,

115 where I = main inlet stream, O = main outlet stream (for both, I/O-OUT is the outflow

116 and I/O-IN is the inflow of the stream). ANTH = anthropogenic outflows, WW =

117 anthropogenic inflows (wastewater), EPT = evapotranspiration, G = groundwater, P =

118 precipitations, and R = runoff.

119 -  $W_I$  refers to the main inlet stream, where returns from the studied compartment

120 can take place. In a marshland, for example, the main inflow ( $W_{I-IN}$ ) is the river, and the

121  $W_I$  outflow ( $W_{I-OUT}$ ) represents infiltrations of saline water into the fluvial system.

122 -  $W_O$  streams represent the main outflow from the system ( $W_{O-OUT}$ ), where the

123 flow can also occur in both directions; thus, returns from the main outflow to the

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3 124 compartment ( $W_{O-IN}$ ) can occur. For example, for an open estuary, these flows represent

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7 125 the tidal, so this main outstream would be subjected to bidirectional flows.

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10 126 -  $W_P$ ,  $W_{EPT}$ ,  $W_G$ , and  $W_R$  are climate and water cycle-related streams that

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13 127 represent the precipitation, the evaporation, the groundwater, and the runoff of the

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15  
16 128 system, respectively. Note that the groundwater flow could also be bidirectional, as it

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19 129 can feed the compartment, but infiltrations from the system to the groundwater network

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22 130 can also occur.

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25 131 -  $W_{ANTH}$  and  $W_{WW}$  are the anthropogenic outflows and inflows in the system,

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28 132 respectively.  $W_{ANTH}$  represents the water taken from the compartment for anthropogenic

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31 133 purposes (for example, for irrigation), and  $W_{WW}$  is the poured water into the system

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34 134 (normally, treated wastewater).

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37 135 Once the streams are identified and quantified, the salt concentration of each flow ( $S_i$ ,

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40 136 kg NaCl /m<sup>3</sup>) is also needed for the calculation of the FF (Eq. (3), in units of time).  $\bar{S}$  is

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43 137 the average salt concentration in the system (kg/m<sup>3</sup>),  $\bar{V}$  the average volume of the

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46 138 studied compartment (m<sup>3</sup>), and  $\bar{F}_i$  the average mass flow of the calculated  $F_{IN}$  and  $F_{OUT}$

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49 139 (kg salt/time) (Eq. (4)), which represent the total mass of salt in the inlet and outlet

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52 140 flows, respectively.  $F_{IN}$  should equal  $F_{OUT}$  as no accumulation takes place and the model

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4 141 is then valid for steady state, but the FF is quantified using  $\overline{F}_i$  (an average) to  
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7 142 acknowledge possible discrepancies of experimental data. Additionally,  $\overline{F}_i$  can be  
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10 143 calculated yearly, monthly, or seasonally, defining the units of the factor itself.  
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$$FF = \frac{\overline{S} \cdot \overline{V}}{\overline{F}_i} \quad (3)$$

$$\overline{F}_i = \sum W_{i-IN} \cdot S_{i-IN} = \sum W_{i-OUT} \cdot S_{i-OUT} = F_{IN} = F_{OUT} \quad (4)$$

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26 145 - **EFFECT FACTOR (EF)**  
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30 146 The EF quantifies the fraction of living species that are going to potentially disappear in  
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33 147 the aquatic ecosystem by the release of a certain chemical<sup>19,27,28</sup>. The available literature  
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36 148 regarding salinity variation effects in aquatic ecosystems models these impacts  
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40 149 according to ecotoxic methodologies, such as USEtox<sup>6,9</sup>. Under that approach, the EF is  
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43 150 calculated employing a species-sensitivity distribution (SSD) curve which represents the  
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47 151 sensitivity of an entire ecosystem to a substance<sup>27</sup>. However, it is constructed based on  
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50 152 the premise that increased exposures will lead to increased effects<sup>19</sup>. Indeed, if the  
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53 153 salinity of a fresh aquatic environment increases, the organisms living in the system will  
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57 154 be negatively affected<sup>10,29-32</sup>. However, negative effects in aquatic ecosystems are also  
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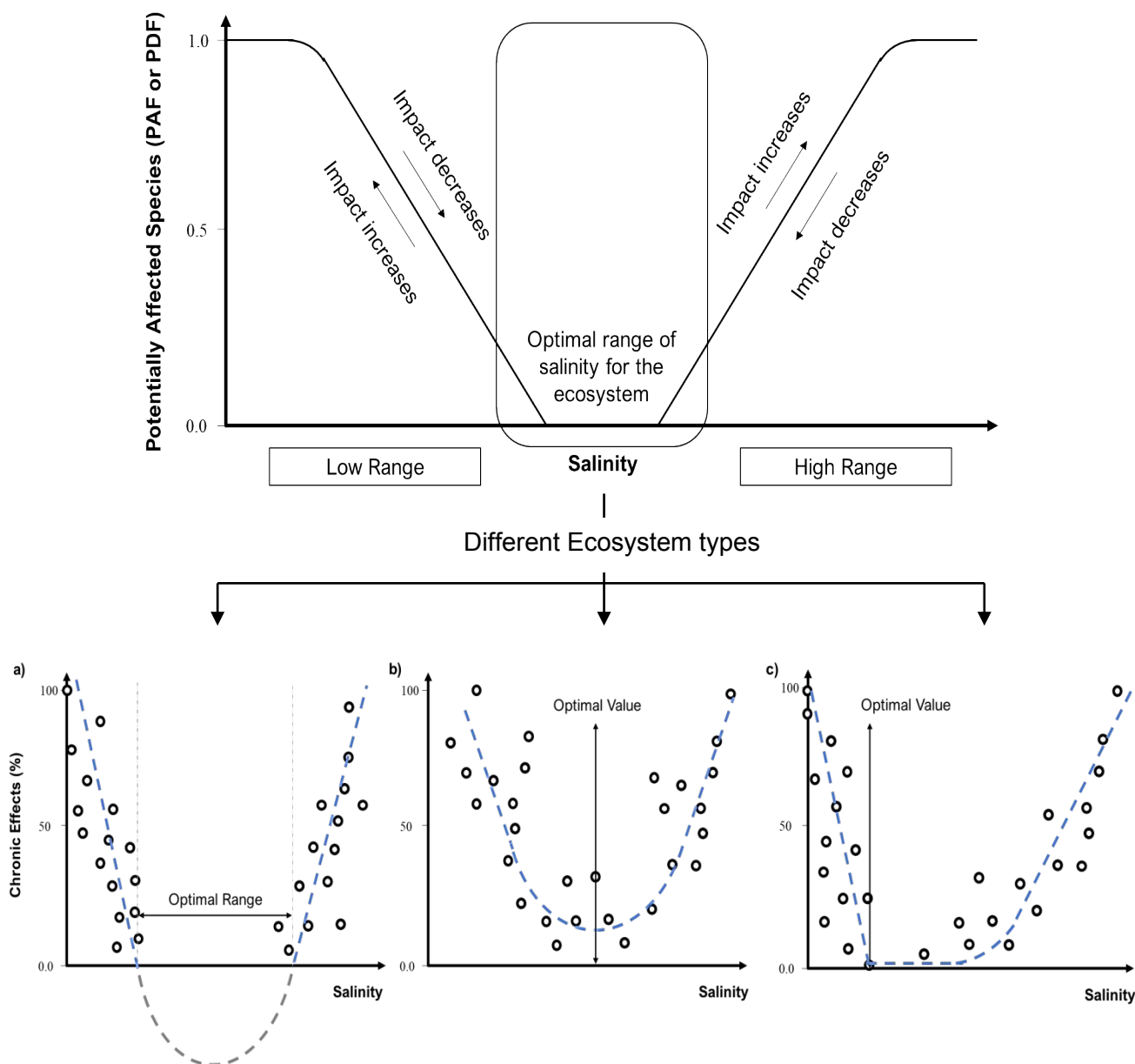
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4 155 reported when salinity decreases, such as massive mortalities reported after exposure to  
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7 156 low salt concentrations due to climatologic events or anthropogenic actions<sup>11,33,34</sup>.  
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10 157 Moreover, some limitations of SSD curves to model ecotoxic impacts due to salinity  
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13 158 have been pointed out<sup>35</sup>, and other indicators, such as species richness, have been  
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17 159 proposed for studies assessing the effects of salinity<sup>31</sup>.  
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21 160 Therefore, variations in the concentration of essential substances can provoke effects in  
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24 161 several directions, as an increase in the salt concentration can be potentially beneficial  
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28 162 for an ecosystem and vice versa, meaning that emission-related impacts can be positive  
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31 163 in some cases. Moreover, currently, it is considered that a substance provokes toxic  
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34 164 effects if it enters in an organism and causes poisoning, endocrine disruption, or other  
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38 165 lethal effects<sup>19</sup>. This approach might be partially accurate for effects of exposure to high  
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41 166 salt concentrations, but it fails when defining the observed effects on ecosystems for  
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44 167 decreases in salinity. This is the reason why the current applied approach (using  
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48 168 ecotoxic methodologies) to model the impacts due to variations in essential substances  
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51 169 (such as salts) need to be reconsidered.  
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55 **170 EFFECT FACTOR FOR SALINITY VARIATIONS IN AQUATIC ENVIRONMENTS**  
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4 171 The proposed approach is based on the premise that the species in an ecosystem have an  
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7 172 optimal range of salinity for living, and that detrimental effects will be observed if it  
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10 173 varies below or above it<sup>36</sup>. Figure 2 shows an ideal representation of a transitional water  
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13 174 body, where there is an optimal concentration of salt at which no impact occurs (i.e.,  
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16 175 PDF = 0). Please note that this figure represents a theoretical hypothesis to be later  
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20 176 verified by the case study application. Then, negative effects will occur if salinity  
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23 177 increases above the optimal range or decreases below it. Moreover, positive impacts  
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26 178 will occur for increments in the salt concentration for environmental concentrations  
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30 179 below the optimal, and vice versa. Note that, for salinity increases in the high range, the  
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33 180 function follows a distribution like the classic SSD curve.

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38 181 The first step is then to define the optimal salt concentration range by gathering data of  
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41 182 chronic effects for the species of the ecosystem regarding salinity (also needed to  
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44 183 quantify the EF itself). If the data are expressed as acute, an acute-to-chronic ratio  
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47 184 (ACR), which is generally  $2^{28}$ , can be used by dividing or multiplying the salt  
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51 185 concentration by the ACR in the high range or in the low range, respectively. Then,  
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54 186 according to our hypothesis (effects will happen above and below a favorable range),  
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4 187 that data representing the chronic effects linked to salinity will be shaped like in the  
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7 188 examples provided in Figures 2a, 2b, or 2c.  
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11 189 For water systems from oligohaline to hyperhaline (see Table S1 at the supporting  
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14 190 information (SI)), it is expected that the concentration-to-response curves have a shape  
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18 191 like the ones shown in Figure 2a or 2b, where the optimal (environmental) salt  
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21 192 concentration can be estimated as a range (the points for which the function has values  $y$   
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24 193  $< 0$ , Figure 2a) or a point (the minimum value of  $y$ , Figure 2b). For freshwater  
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28 194 environments, it is more likely that the impacts in the ecosystem are due to salt  
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31 195 concentration increases. For the part of the curve representing salinization, a classic  
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34 196 SSD approach might be a fair approximation. Nevertheless, the whole system profile is  
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38 197 only provided if both ranges (high and low) are included, where the optimal  
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41 198 (environmental) concentration can be found as the intersection of the two functions (a  
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45 199 lineal one for the low range and SSD-like for the high range, Figure 2c).  
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201 **Figure 2.** Expected (hypothetical) shape of the curve defining the optimal salinity range  
 202 for aquatic ecosystems. The main plot is an ideal representation of the impacts of salt  
 203 variations, and the graphs a, b, and c are theoretical representations of different types of  
 204 ecosystems, where a and b shape the impacts of transitional or non-fresh compartments,  
 205 and c is a freshwater system.

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4 206 After defining the optimal region, there are two EFs:  $EF_{LOW}$  and  $EF_{HIGH}$ , both in  
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7 207  $PDF \cdot m^3/kg$ . They represent the amount of a substance that generates a certain effect on  
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10 208 the ecosystem, where the EF is the slope of the concentration-response curve.  $HC50_{LOW}$   
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13 209 and  $HC50_{HIGH}$  (both in  $kg NaCl/m^3$ ) represent the salt concentration that generates an  
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17 210 effect in 50% of the ecosystem species in the low and high range, respectively; thus, for  
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20 211 effects in 50% of the species, EFs are expressed as in Eq. (5) and Eq. (6). HC50s are the  
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23 212 geometric mean of the individual species  $EC50^{28}$ , which is the concentration of  
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27 213 pollutant that generates an effect in the 50% of the individuals of a single species. In  
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30 214 this sense, an SSD-like approach is still maintained.

$$EF_{LOW} = \frac{0.5}{HC50_{LOW}} \quad (5)$$

$$EF_{HIGH} = \frac{0.5}{HC50_{HIGH}} \quad (6)$$

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43 215 Assuming a classic SSD-like curve (i.e., log-logistic functions), EC50s could be  
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46 216 calculated using to mirrored sigmoidal curves. Moreover, according to our hypothesis  
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49 217 (Figure 2), EC50s could be estimated by fitting the chronic effect data to a quadratic  
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53 218 function (see Figure S1 at the SI). Note that the log-logistic and the quadratic curves  
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56 219 only converge at intermediate ranges of effects, so this approximation might not be  
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59 220 accurate to calculate EFs based on HC10 or HC20, as new trends on ecotoxicity  
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4 221 modeling suggest the use of EC10s or EC20s to model the factors<sup>19</sup>. Finally, the sign of  
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7 222 each EF will be established depending on the direction of the change: increasing the salt  
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10 223 concentration in the low range accounts for a negative  $EF_{LOW}$  (impacts decline), and  
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13 224 decreasing it accounts for a positive  $EF_{LOW}$  (impacts increase), and vice versa for the  
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17 225 high range.

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## 34 229 APPLICATION OF THE METHODOLOGY TO A CASE STUDY

### 39 230 - FATE FACTOR CALCULATION

43 231 For the case of *Arousa ría*, Figure 1 is simplified:  $W_G$  is neglected due to the  
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45  
46 232 insignificant contribution of the groundwater to the total flow of the *ría*<sup>37</sup>, and  $W_{I-OUT}$  is  
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49 233 ignored since no infiltrations from the estuary to the river are expected (see Figure S2).  
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52  
53 234 The remaining streams are quantified in cubic meters per month, covering from 2011 to  
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55  
56 235 2018 according to the data availability, as detailed bellow. First, stream flows are  
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60 236 determined:

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4 237 -  $W_{I-IN}$  corresponds to the flows of rivers Ulla and Umia, which are monthly  
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7 238 measured and reported by local administrations<sup>38</sup> in several locations along the river  
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9  
10 239 courses, so the closest point to the *ría* was chosen.

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12  
13 240 -  $W_{WW}$  can be divided into fresh wastewater ( $W_{WW-FRESH}$ ) and salty one ( $W_{WW-}$   
14  
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16  
17 241  $SALT$ ). It was estimated that the area of *Arousa ría* is responsible for about 6.6% of the  
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19  
20 242 total ( $W_{WW}$ ) Galician discharges<sup>37,39</sup> and that 82–92% of this amount (yearly variation  
21  
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23  
24 243 in 2011-2018) corresponded to fish-canning wastewater<sup>37,39</sup>, the latter representing  
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26  
27 244 virtually all saline emitting sectors ( $W_{WW-SALT}$ , in cubic meters per year). Then, to  
28  
29  
30 245 obtain monthly data, a distribution was defined based on the shellfish and seafood  
31  
32  
33  
34 246 harvesting and processing seasonal pattern to estimate  $W_{WW-SALT}$  in  $m^3/month$  (see  
35  
36  
37 247 Table S2). On the other hand,  $W_{WW-FRESH}$  (including urban wastewater and fresh  
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39  
40 248 industrial streams, in cubic meters per year) was estimated as the subtraction of  $W_{WW-}$   
41  
42  
43  
44 249  $SALT$  from the total  $W_{WW}$ . Then, to obtain monthly data, it was assumed that fresh  
45  
46  
47 250 discharges are evenly distributed along the months of the year (also considering that  
48  
49  
50 251 freshwater streams represent a flow considerably lower than salty streams).

52  
53 252 -  $W_{ANTH}$  is not linked here to irrigation, as estuary water has high salinity and  
54  
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56  
57 253 Galicia is an area with high precipitations, but the local fish-canning industries use  
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4 254 seawater for their processes with variable flows. Due to the lack of more precise data,  
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6  
7 255 we assumed that 75% of water input for fish-canning processes was saline water and  
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9  
10 256 25% was freshwater<sup>40-43</sup> and that there are no water losses in the process, i.e., inputs =  
11  
12  
13  
14 257 outputs, thus  $W_{ANTH} = 0.75 \cdot W_{WW-SALT}$  .  
15  
16  
17 258 - Evaporation rates were estimated using empirical correlations as indicated in Eq.  
18  
19  
20 259 (7)<sup>44,45</sup>, where  $W_{EPT}^*$  is stated in L/m<sup>2</sup>·time (i.e., monthly averages in this case),  $u_2$  is the  
21  
22  
23 260 wind speed (m/s),  $A$  is the evaporating area (m<sup>2</sup>),  $v_w^*$  is the saturated vapor pressure at  
24  
25  
26  
27 261 the water surface temperature (kPa), and  $v_a$  is the partial vapor pressure in the air at 2 m  
28  
29  
30 262 height (kPa) calculated by Eq. (8), where  $v_a^*$  is the saturated vapor pressure at air  
31  
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33  
34 263 temperature (kPa) and  $\phi$  is the relative humidity (%). Finally, to obtain  $W_{EPT}$  in  
35  
36  
37 264 equivalent units to the rest of the streams (m<sup>3</sup>/month), Eq. (9) is needed.

$$W_{EPT}^* = (2.36 + 1.72 \cdot u_2) \cdot A^{-0.05} \cdot (v_w^* - v_a) \quad (7)$$

$$v_a = v_a^* \cdot \phi \quad (8)$$

$$W_{EPT} = \frac{W_{EPT}^* \cdot A}{1000} \quad (9)$$

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51 265 - Average monthly precipitations ( $W_p$ ) were obtained from the meteorological  
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53  
54 266 station located in Corón (Vilanova de Arousa)<sup>46</sup>.

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4 267 -  $W_R$  (runoff) was taken as the average values of the South West basins of  
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6  
7 268 Galicia<sup>47</sup>. Being the only data set available, the monthly distribution and expected runoff  
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9  
10 269 flow for 2002 were applied to the 2011-2018 period defined.  
11  
12  
13  
14 270 At this point, all the flows, except  $W_{O-IN}$  and  $W_{O-OUT}$ , which represent the tidal, are  
15  
16  
17  
18 271 quantified, so mass balances must be solved. First, Eq. (10) and Eq. (11) present the  
19  
20  
21 272 general steady state water and the salt balances, respectively, where  $S_i$  is the salt  
22  
23  
24 273 concentration ( $\text{kg NaCl/m}^3$ ) of each stream  $W_i$ .

$$W_{O-IN} + W_I + W_R + W_P + W_{WW} = W_{O-OUT} + W_{EPT} + W_{ANTH} \quad (10)$$

$$W_{O-IN} \cdot S_{O-IN} + W_I \cdot S_I + W_R \cdot S_R + W_P \cdot S_P + W_{WW} \cdot S_{WW} = W_{O-OUT} \cdot S_{O-OUT} + W_{ANTH} \cdot S_{ANTH} \quad (11)$$

274 Note that Eq. (10) could be written as  $W_{IN} = W_{OUT}$ , representing the total inflows and  
275 outflows in the estuary. Then, to accurately quantify the tidal streams, new mass  
276 balances will be stated, representing a system as natural as possible (Eq. (12) and Eq.  
277 (13)), where no anthropogenic activity exists:

$$W_{O-IN} + W_I + W_R + W_P = W_{O-OUT} + W_{EPT} \quad (12)$$

$$W_{O-IN} \cdot S_{O-IN} + W_I \cdot S_I + W_R \cdot S_R + W_P \cdot S_P = W_{O-OUT} \cdot S_{O-OUT} \quad (13)$$

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4 278 To be able to proceed, the salt concentration of each stream,  $S_i$ , needs now to be  
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7 279 quantified:  
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11 280 - The salt concentration of the ocean ( $S_{O-IN}$ ) was considered constant at 36 g  
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13  
14 281 NaCl/L, as it is the average estimated sea surface salinity measured between 2004 and  
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17  
18 282 2013 for this Atlantic area by satellite monitoring<sup>4</sup>.  
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20  
21 283 - The salt concentration of the estuary ( $S_{O-OUT}$ ,  $S_{ANTH}$ , but also  $\bar{S}$  in Eq. (3)) is  
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23  
24 284 variable according to the spatial distribution and the tidal intensity. Water is fresher near  
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27  
28 285 the river mouth and saltier near the ocean, but the salt concentration also increases with  
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30  
31 286 depth because salty water has a higher density<sup>48</sup>. Data collected from two buoys  
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34 287 (*Ribeira*, in an intermediate location between the river mouth and the ocean, and  
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36  
37 288 *Cortegada*, near the river mouth; see map in Figure S3)<sup>46</sup> were used to determine the  
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40  
41 289 monthly average salinity of the estuary.  
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44  
45 290 - Galician rivers are not salinized, presenting low salinity and conductivity<sup>49</sup>.  
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47  
48 291 Normally, chloride concentrations of Spanish rivers range between 0.010 and 0.030 kg  
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50  
51 292 Cl/m<sup>3</sup>, and recent studies for Galician rivers near shore areas reported values of 0.0103  
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54  
55 293 – 0.0222 kg Cl/m<sup>3</sup><sup>50</sup>. An average value of 0.020 kg NaCl/m<sup>3</sup> is considered for  $S_I$  and  
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57  
58 294  $S_R$ .  
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4 295 - Chloride concentration in rainwater is generally very low, although it varies  
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7 296 worldwide depending on wind intensity and seawater proximity. Measurements of the  
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10 297 chloride concentration in rainwater in Spain and Portugal averaged 0.020 kg Cl/m<sup>3</sup>, and  
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13 298 the value reported for the Galician station, located at around 40 km from the coast, was  
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17 299 0.003 kg Cl/m<sup>3</sup><sup>51</sup>. A slightly greater salinity of 0.005 kg NaCl/m<sup>3</sup> is applied for S<sub>P</sub> to  
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20 300 consider the effect of sea salt aerosols.  
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23 301 - Eq. (14) and Eq. (15) are used to calculate S<sub>WW</sub>, where W<sub>WW-SALT</sub> is the yearly  
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25  
26 302 flow of industrial fish canning wastewater discharged into the estuary (m<sup>3</sup>/year), and  $\bar{S}_t$   
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28  
29 303 is the average salt concentration of the estuary each year (kg/m<sup>3</sup>). It was assumed that  
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33 304 S<sub>WW-FRESH</sub> coming from drinkable sources was 0.5 kg NaCl/m<sup>3</sup><sup>52</sup>, and that S<sub>WW-SALT</sub>  
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35  
36 305 contained 75% of estuarine water and 25% of fresh (drinkable) water as previously  
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39  
40 306 indicated. The salinity of waste streams poured into the *ría* S<sub>WW</sub> can be estimated as an  
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42  
43  
44 307 average value as shown in Eq. (15).  
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46  
47

$$S_{WW-SALT} = \frac{0.75 \cdot W_{WW-SALT} \cdot \bar{S}_t + 0.25 \cdot W_{WW-FRESH} \cdot 0.5}{W_{WW-SALT}} \quad (14)$$

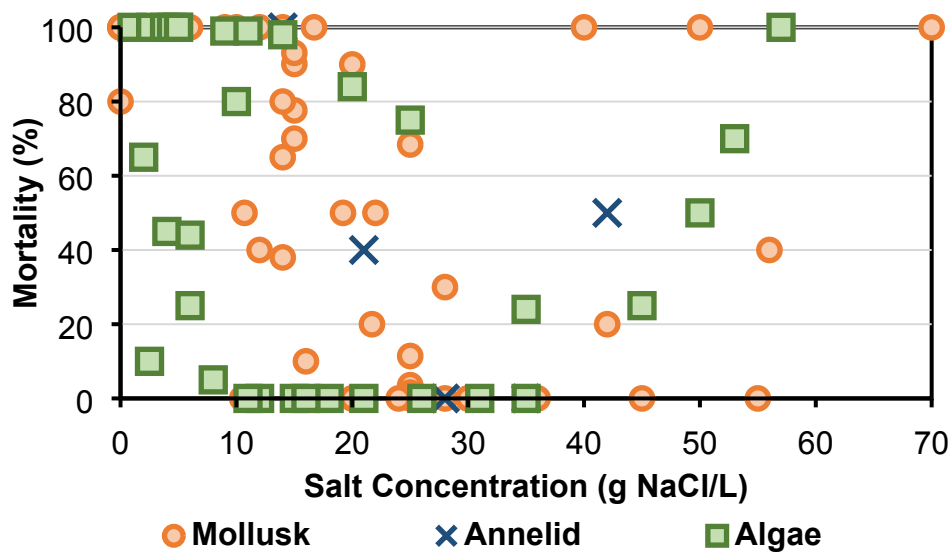
$$S_{WW} = \frac{W_{WW-SALT} \cdot S_{WW-SALT} + W_{WW-FRESH} \cdot S_{WW-FRESH}}{W_{WW-SALT} + W_{WW-FRESH}} \quad (15)$$

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4 308 Finally, the FF can be calculated. As already stated, both monthly and yearly results can  
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7 309 be obtained, as well as seasonal values, the dry season spanning June, July and August  
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10 310 in the case study area, and the remaining months are the wet season. To see detailed  
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13 311 monthly/seasonal/yearly results and other detailed information about the calculation  
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17 312 procedures, see the Excel file and the Section SIII in the SI (see Table S3).  
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21 313 - **EFFECT FACTOR CALCULATION**  
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25 314 To calculate the EFs, data of chronic effects were gathered (for this study, only  
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28 315 mortality was considered). Then, the collected data were represented together to find the  
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32 316 ecosystem optimal (environmental) salt concentration (Figure 3). Note that this value is  
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36 317 not going to be employed in any other further calculation, and it is just used to  
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39 318 determine which data will be utilized to generate  $EF_{LOW}$  and  $EF_{HIGH}$ . In this sense,  
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42 319 several fittings could be tested, as shown in Figure 2a and 2b. Different approaches  
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46 320 were applied, evaluated, and discussed (see Section SIV of the SI, and Figures from S4  
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48  
49 321 to S7) to identify this cutoff point. These strategies pointed out a possible environmental  
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52 322 optimal range of 24–36 g NaCl/L, where the average point obtained after testing  
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56 323 different fittings is  $31.9 \pm 1.4$ , which will be the cutoff to define the low and high ranges  
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59 324 of salt concentration comprising  $EF_{LOW}$  and  $EF_{HIGH}$ , respectively.  
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4 325 Therefore, EC50 concentrations are calculated for each species at high and low range  
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7 326 considering the estimated cutoff point. Most of the information gathered referred to  
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10 327 effects at low range, so the EC50<sub>LOW</sub> for these species can be obtained directly using a  
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13 328 sigmoidal curve with a profile mirrored to the classic SSD curve fittings. However,  
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17 329 EC50<sub>LOW</sub> and EC50<sub>HIGH</sub> can be also estimated for each species by using the quadratic  
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20 330 approach of our hypothesis, which provides effects at both ranges (as in Figure S1; see  
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24 331 Table S4 for detailed information of how the quadratic approximation was used).



332  
333 **Figure 3.** Distribution of the data regarding mortality of different species present in  
334 *Arousa ría*. The data used to generate this plot are specified in Table 1 and Table S4 and  
335 Section SIV of the SI.



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4 336 The value of each EC50 (high and low) and both HC50s are shown in Table 1. The  
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7 337 numbers in italic indicate that the concentration was obtained by the quadratic fitting  
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10 338 using data from the low range of salinity (for *V. senegalensis* and *S. polychides*, the  
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13 339 power function had a convex profile, so the data were estimated using the inverse  
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16 340 function, see Table S4). The results of EF<sub>HIGH</sub> and EF<sub>LOW</sub> (Table 1) fit with the expected  
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20 341 system behavior, as biotic diversity in estuaries starts to decline above a salinity of  
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22  
23 342 about 40 kg NaCl/m<sup>3</sup>, with most species unable to survive in salinities above 50 kg  
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27 343 NaCl/m<sup>3</sup> <sup>36</sup>.

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31 344 - **CHARACTERIZATION FACTOR CALCULATION**  
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35 345 CFs were obtained according to Eq. (2). Here, the EF achieves different values  
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38 346 according to the level of salinity (high and low), while the FF, which is supposed to be  
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42 347 the same for both ranges of salinity (the water streams will have the same physical  
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45 348 distribution regardless of the salt concentration), varies seasonally (wet/dry season).  
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49 349 Therefore, there are six possible CFs to use (Table 1).  
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53 350 **Table 1.** Fate Factors, Effect Factors and Characterization factors modeling the effects  
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57 351 of salinity variations in *Arousa ría*. For the EF section, the EC50s in italic represent the  
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59  
60 352 values estimated assuming quadratic distribution (data for chronic effects were only

353 available for low range). The results are expressed as average  $\pm$  standard deviation and  
 354 the confidence interval are between brackets, where negative results were assumed zero.

Fate Factors (month/year)					
Wet (month)		Dry (month)		Annual (year)	
6.84 $\pm$ 1.84		4.02 $\pm$ 1.52		4.51 $\pm$ 1.51	
[1.32, 12.36]		[0, 8.59]		[0, 9.05]	
Effect Factors, HC50s and EC50s					
Species			EC50 <sub>LOW</sub>	EC50 <sub>HIGH</sub>	
<i>Ruditapes philipinarium</i> (Japanese clam) <sup>11,53</sup>			16.0	34.4	
<i>Ruditapes decussatus</i> (Grooved carpet shell) <sup>11,54</sup>			15.5	42.9	
<i>Cerastoderma edule</i> (Common cockle) <sup>11,55</sup>			20.4	61.7	
<i>Vanerupis corrugata</i> (Pullet carpet shell) <sup>11,53</sup>			21.7	37.8	
<i>Saccharina latissima</i> (Sea belt, brown algae) <sup>56,57</sup>			8.6	39.1	
<i>Diopatra neapolitana</i> (Polychaete) <sup>58</sup>			19.0	42.3	
<i>Donax trunculus</i> (Wedge clam) <sup>59</sup>			19.3	32.1	
<i>Mytilus galloprovincialis</i> (Mediterranean Mussel) <sup>60</sup>			9.8	38.2	
<i>Scrobicularia plana</i> (Peppery furrow shell) <sup>55</sup>			10.7	40.4	
<i>Saccorhiza polyschides</i> (Furbellow, brown algae) <sup>61</sup>			30.1	40.1	
<i>Zostera noltei</i> (Dwarf eelgrass, seagrass) <sup>62,63</sup>			1.3	49.6	
<b>HC50 (Geometric mean)</b>			<b>12.7</b>	<b>41.1</b>	
HC50 arithmetic mean			15.7 $\pm$ 7.8	41.7 $\pm$ 8.0	
Effect Factors (PDF·m <sup>3</sup> /kg)					
EF <sub>LOW</sub>			EF <sub>HIGH</sub>		
0.04 $\pm$ 0.02			0.01 $\pm$ 0.002		
[0, 0.1]			[0.005, 0.02]		
Characterization Factors (PDF·month·m <sup>3</sup> /kg)					
CF <sub>LOW</sub>			CF <sub>HIGH</sub>		
Dry	Wet	Annual	Dry	Wet	Annual
0.27 $\pm$ 0.21	0.16 $\pm$ 0.14	0.18 $\pm$ 0.15	0.08 $\pm$ 0.04	0.05 $\pm$ 0.03	0.05 $\pm$ 0.03
[0, 0.89]	[0, 0.57]	[0, 0.62]	[0, 0.20]	[0, 0.13]	[0, 0.14]

355 For the obtained FFs, the standard deviation acknowledges the discrepancy of steady  
 356 state assumption (see Section SIII of the SI). For the EF, the confidence intervals and

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3 357 the standard deviation of the factor were calculated by using the arithmetic average (see  
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7 358 Table 1). For all factors, the uncertainty intervals were obtained considering three times  
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10 359 the standard deviation, and negative lower bounds were considered zero.  
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14 360 CFs are reported as absolute values, but they depend on the direction of the change (i.e.,  
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18 361 increase or decrease in salt concentration at low or high range). Therefore, to quantify  
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21 362 the effects of a dam release during the wet season which provokes a decrease in the salt  
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24 363 concentration,  $CF = 0.16 \text{ PDF}\cdot\text{month}\cdot\text{m}^3/\text{kg}$  (i.e. impact increases); but to quantify the  
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27  
28 364 impact of a saline effluent discharge in the same situation (wet season, low salinity),  $CF$   
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31 365  $= -0.16 \text{ PDF}\cdot\text{month}\cdot\text{m}^3/\text{kg}$  should be used as salinity is already below the optimal, so the  
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35 366 increase in salt concentration has a positive effect in the receiving waters. In this  
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38 367 example,  $CF_{\text{LOW}}$  is used instead of  $CF_{\text{HIGH}}$  because *Arousa ría* has a salt concentration  
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41 368 below the optimal<sup>49</sup>.  
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## 45 369 **DISCUSSION**

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50 370 When evaluating the effects of pollutants' release, it is logic to assume that a rise in the  
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53 371 chemical concentration generates an increase in the impacts. However, anthropogenic  
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56 372 activities are affecting the planetary biogeochemical cycles, and impacts are now not  
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59 373 only due to the release of harmful substances, but also to variations in the environmental  
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3 374 conditions of ecosystems provoked by unnatural changes. There are some substances  
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7 375 (e.g., salt, nitrogen, dissolved oxygen) to which the classic ecotoxic impact approach  
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10 376 might just fit partially. Indeed, a drastic increase in the substance concentration will  
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13 377 generate adverse impacts. However, at the same time, a certain concentration in the  
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17 378 media is needed to support the ecosystem survival.

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21 379 In recent years, concern for salt variations in the different environmental compartments  
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24 380 has increased, but the models available to describe their impacts are still scarce and the  
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27  
28 381 current literature uses ecotoxicity models to assess these effects<sup>6,12,18,64</sup>. For instance, the  
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31 382 USEtox model, one of the most used methods to evaluate ecotoxic impacts in LCA, has  
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34 383 not addressed effects of salt releases so far<sup>12</sup> and does not include the coastal seawater  
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38 384 and brackish areas as environmental compartments<sup>19</sup>. Therefore, new approaches are  
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41 385 necessary to include essential substances and these critical areas in the impact  
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45 386 assessment models, and specific methodological choices must be implemented to  
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48 387 develop CFs for essential elements in non-freshwater environments.

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52 388 Regarding the FF, it is expressed in this work in units of time (see the fate factor section  
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56 389 in materials and methods). This provides a suitable framework to model the fate of  
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59 390 essential substances. The few research works that have assessed the effects of salinity  
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4 391 variations in aquatic environments solved the FF question from different approaches. In  
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7 392 a first study, when evaluating the impacts of brine disposal<sup>9</sup>, the different elements  
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10 393 present in brine were grouped. NaCl had a high residence time, so the FF was chosen as  
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12  
13 394 the residence time of the second most persistent element of the salinity group ( $\text{Cu}^{2+}$ , 37  
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17 395 days). However, this approximation can have high errors due to the difference in the  
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19  
20 396 elements' residence times (from days to millions of years). In another study, when  
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23 397 steady-state mass balances were applied to the water streams and the salt in a coastal  
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27 398 wetland<sup>6</sup>, the units of the FF were g·year/L, yielding in a dimension of time per  
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29  
30 399 concentration. This factor was useful to directly link salt variations in the wetland not  
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33 400 only with ecotoxic impacts, but also with social and economic effects as crop loss.  
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37 401 However, the unit of that FF complicates the comparison of the results obtained there  
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40 402 with the present study. Moreover, it hinders the evaluation of the impacts of salinity  
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43 403 variations on the ecosystem linked to different impact categories as ecotoxicity.  
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47 404 Therefore, to remain coherent and consistent with other impact categories, it is  
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50 405 necessary to provide factors that align with the standard units of the impact assessment  
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53 406 stage and that are used by other methodologies recommended by the UNEP's Life  
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57 407 Cycle Initiative, such as the USEtox. This harmonization aspect also applies to the  
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3 408 measure of the EF, which is PDF in this study because UNEP's Life Cycle Initiative  
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7 409 recommends to base ecosystem damage estimates on this metric<sup>19,65</sup>.  
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11 410 As for the EF, the proposed approach copes with the fact that impacts might be due to  
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14 411 increases but also decreases in the concentration of essential substances, and it  
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17 412 acknowledges potential benefits linked to emission-related impacts. In this sense, a  
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21 413 classic SSD-based methodology (typically used in ecotoxicity) appears as a practical  
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24 414 and useful tool to predict the possible negative effects of a pollutant's release on  
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27 415 ecosystems. However, although some of the limitations linked to the use of SSD curves  
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31 416 have been pointed out in the past few years, the principles shaping the methodology  
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34 417 have remained unchanged for decades<sup>35</sup>. Additionally, the use of classic ecotoxicity  
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37 418 methodologies also have some limitations to model impacts due to variations in the  
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41 419 concentration of essential substances, since the effects of these variations do not fit the  
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44 420 classic definition of toxicity (linked to poisoning, endocrine disruption, etc.).  
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49 421 For the present study, a quadratic function was used to shape the effects of salinity on  
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52 422 ecosystems instead of the typical log-normal or log-logistic distribution applied in the  
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55 423 SSD-based methods, by applying this approximation for the calculation of EC50s (and,  
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58 424 therefore, the EFs). Nevertheless, log-logistic distributions were also applied to the  
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3 425 chronic data gathered for each individual species to test the robustness of the  
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7 426 approximation. The differences between the EC50s found applying log-logistic and  
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10 427 quadratic fittings averaged 3.82% (data not shown), meaning that the quadratic  
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13 428 approach is accurate to estimate effects at intermediate ranges of concentration in an  
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17 429 LCA context (but might not be accurate for EC10s or EC20s). This approximation was  
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20 430 used to estimate the EC50s and HC50s at both concentration ranges (low and high), but  
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23 431 the SSD-based methodology fundamentals are maintained. Thus, the current study does  
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27 432 not aim to question the SSD curves themselves, but to expand their scope to understand  
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30 433 new ecological features.

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34 434 In fact, this quadratic approximation can be especially useful for systems from  
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38 435 oligohaline to hyperhaline (see Table S1), but it might not be as accurate for freshwater  
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41 436 systems. Here, the expected effects are linked to salinization, while impacts linked to a  
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45 437 sharp freshening are unlikely. In fact, the most important impacts derived from  
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48 438 anthropogenic activities and climate change in freshwater ecosystems are expected to be  
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51 439 linked to salinization<sup>16</sup>, where the SSD-based methodology can be a fair approximation  
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55 440 to determine the effects of salinity increase if the effects of freshening are neglected.  
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4 441 The truth is that the few studies considering the effects of salt variations in aquatic  
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7 442 environments considered SSD approaches. In a wetland-related study<sup>6</sup>, an SSD curve  
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10 443 was constructed considering that anthropogenic activities (irrigation) were provoking  
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13 444 salinization, which seems to be a fair approximation considering that the salt  
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17 445 concentration in the water body had increased from 2.6 g/L in 1983 to 7.50 g/L in 2008.  
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20 446 However, the salinity conditions of the wetland were oligohaline, so impacts related to  
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23 447 freshening could also take place (due to rainfalls, for example). Therefore, the  
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27 448 approximation was fair to evaluate salinization, but failed at evaluating the effects due  
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30 449 to potential freshening.  
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34 450 In a brine disposal research work<sup>9</sup>, a concentration of 40 kg NaCl/m<sup>3</sup> was chosen as  
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38 451 EC50. Analogously to the wetland case, this approximation might be fair considering  
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41 452 that desalination plants would discharge their briny effluents in marine waters, and the  
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45 453 effects of this saline disposal are expected to be negative impacts linked to salinity  
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48 454 increase. However, the modeling of the system is still not fully comprehensive if the  
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51 455 low ranges of salinity are no included. In fact, a case where a transitional brackish  
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55 456 ecosystem is subjected to freshening and where brine disposal is potentially beneficial  
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58 457 could take place under this point of view (impacts would decrease due to brine  
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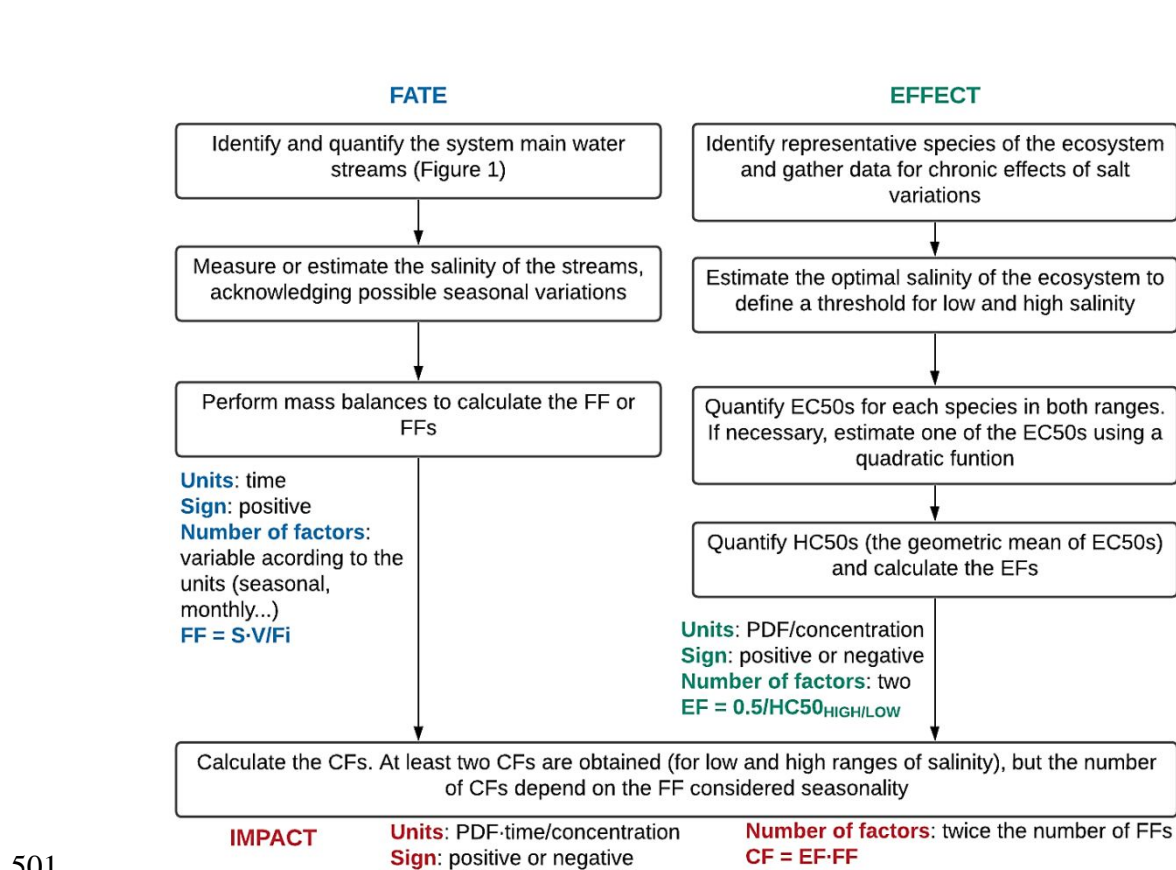
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3 458 disposal), so an approach where this is not considered is lacking some relevant  
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7 459 information for the system.  
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11 460 Finally, these divergent approaches hinder a discussion of the obtained CF. The wetland  
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14 461 and brine CFs, in terms of potentially affected fraction of species (PAF), were 0.32  
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17 462 PAF·yr and 0.47 PAF·m<sup>3</sup>·day/kg, respectively, which are similar to the ranges obtained  
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21 463 here (0.05 – 0.27 PDF·m<sup>3</sup>·day/kg). However, for the wetland, the units are not  
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24 464 comparable, and, for both cases, the obtained CFs were at the midpoint level (measuring  
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27 465 PAF, not PDF), so the discussion is not straightforward. Nevertheless, it is important to  
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31 466 point out how the quantification of CFs to measure the effects of salinity variations can  
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34 467 be relevant for the management of anthropogenic activities in sensitive ecological areas,  
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37 468 such as transitional waters. In fact, the method described and applied here has the  
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41 469 potential to support decision-making processes around effluent discharge, industrial  
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44 470 stream management, brine disposal control, and dam flow regulation, by providing  
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47 471 useful information about when and how to discharge these anthropogenic streams with s  
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51 472 minimum or even a positive impact.  
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55 473 To apply the CFs developed here, the life cycle inventory (LCI) shall record the mass of  
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59 474 salts released to the aquatic environment per functional unit, acknowledging possible  
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4 475 differences between the wet and the dry season when relevant and if information is  
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11 477 **FUTURE OUTLOOK**

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15 478 The presented novel approach proposes a model to shape the effects of variations in the  
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18 479 concentration of an essential substance in the environment (Figure 4). Here, the effects  
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22 480 in the ecosystem might not be always directly proportional to the pollutant  
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25 481 concentration. Although the methodology was applied to salt (i.e., NaCl), the same  
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28 482 principles can be implemented for other essential substances such as macronutrients,  
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32 483 other salts, metals, and even resources such as water. Moreover, as the definition of  
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35 484 salinity is broader than just sodium chloride, a comprehensive salinity assessment may  
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38 485 shape the effects of varying the concentration of other substances. Furthermore, by  
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42 486 including elements such as carbonate, nitrate and sulfate, a comprehensive study of the  
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45 487 ecosystem salinity might extend the effects of the observed variations to other  
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48 488 environmental categories (such as climate change, eutrophication, or acidification,  
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52 489 respectively). In any case, the uniformization of the CFs for salinity impacts (expressed  
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55 490 in the consensus units) opens a new pathway where the effects due to variations in the  
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58 491 concentration of essential substances can be fully assessed for the first time.

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4 492 This pathway has a clear bottleneck, which might hinder its extensive application, and  
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7 493 which is not new to the LCA community, i.e., regarding data availability. Fortunately,  
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10 494 the access to information is becoming easier as science is becoming more accessible and  
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13 495 data is being acquired through more sophisticated means, such as dedicated satellites,  
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17 496 which can be more easily managed through advanced computational methods. In this  
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20 497 sense, although the provided CF only has local applications, the methodological  
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23 498 approach is transferable to any other region. Although some data might be difficult to  
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27 499 find, a preliminary guidance for data acquirement in the CF development and  
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30 500 application is provided in Section SV of the SI.  
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501  
502 **Figure 4.** Overview of the steps that must be followed to apply the methodology to  
503 assess the effects of salinity variations in aquatic environments.

#### 504 SUPPORTING INFORMATION

505 The supporting Word file contains additional information about aquatic environments  
506 regarding salinity, about the application of the quadratic approximation for the EF, and  
507 about the calculation of the FF. The supporting Excel file contains all the calculations  
508 performed to quantify the CF.

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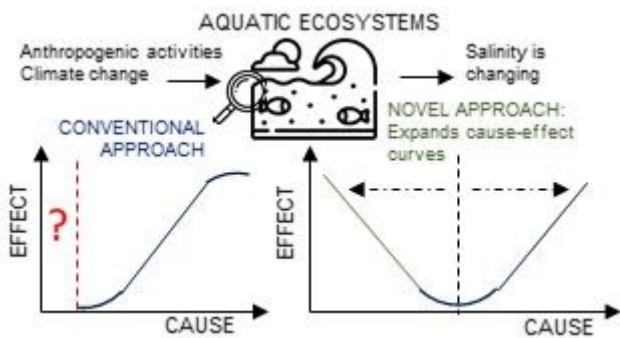
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