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INTERRUPTED CASE STUDY: TOXI-Cd FISHES IN THE DAM

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Part I: COM A ÁGUA NA BOCA

The Nebula Reservoir is an artificial water reservoir created to generate electricity and supply water to the city of Greenfields, 20 km away. The reservoir has this name due to the region flooded by the Spring River. With approximately 15 km in length, 500 m in width and a depth that varies between 2 and 15 meters, the waters of Nebula Reservoir are also used for irrigation, watering of animals, fishing, spa and leisure activities, such as water skiing and boat trips.

Thomas is a graduate student in Chemistry at the Greenfields University. He now lives in Greenfields but regularly visits his parents in the reservoir area. It was late afternoon in 2013 when Thomas arrived home and greeted his mother, Jacinta.

- Hi Mom! How are you?

- Oh, my son! Everything is good! And with you?

- I am well too. I felt like having that fantastic fish you make, so I decided to stop by.

Like the others in the Nebula region, Thomas's family has a diet based on fish caught in the dam. However, that night, it would not be possible for Thomas to taste his mother's dish.

- I think you'll have to go without then.

- Why's that? - Asks Thomas indignantly.

- Do you know Rita, our neighbor? She has been feeling leg pain, stomachache, diarrhea and vomiting. She did some tests and discovered she was experiencing zinc and cadmium intoxication.

- How did this happen? Is she better? What does this have to do with the fish?

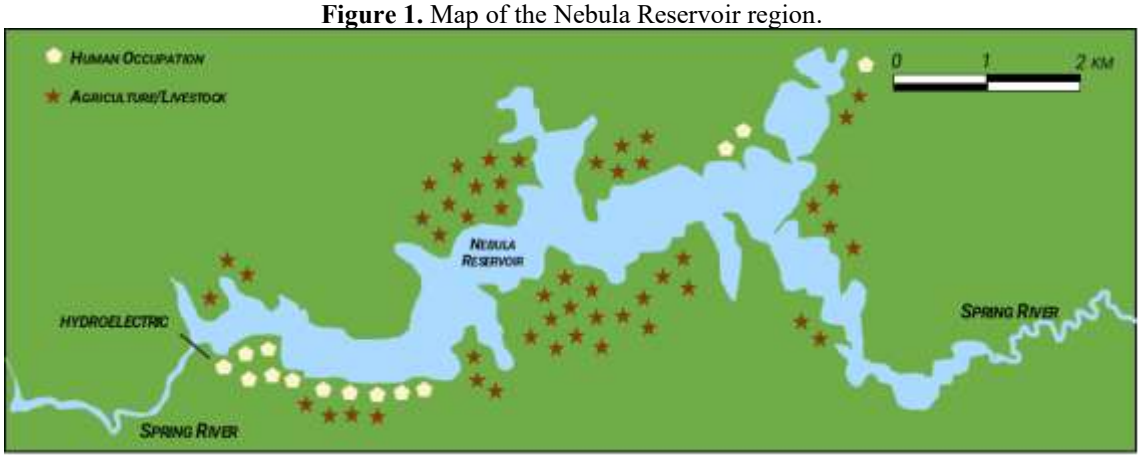
- She's fine now. The doctor told her to stop eating the fish in the dam, as he believes the water is contaminated with metals. I heard about other people in the region who also experienced the same symptoms. So, at home, we are taking a break from the fish.

- Wow, what a serious problem! Since there won't be any fish, I'll do some research and try to find what could be causing all this.

- It would be great to know what is happening with our dam. Next week, I'll go to the market, buy a fish and make it for you, okay?

While his mother prepared dinner, Thomas researched the Nebula Reservoir region on his cell phone and found the following map.

Figure 1. Map of the Nebula Reservoir region.



Activities – Part I

1. What do you already know about the case study situation? Have you read about similar situations? What experiences related to the subject addressed in this case have you had previously?

2. Based on the case narrative, **formulate hypotheses** about the origin of the cadmium and zinc contamination of the Nebula region inhabitants and justify the relevance of these hypotheses.

3. Based on the case narrative, **propose a question** for your group to investigate.

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Part II – Around the Nebula Reservoir

Upon returning home, Thomas continued the research he was carrying out and found some more information about the Nebula Reservoir and the activities carried out around it.

"The Nebula Reservoir is responsible for 40% of Greenfields' water supply, with the rest being collected directly from the Spring River in an area closer to the urban perimeter. In a study carried out by the Greenfields' Water and Waste Department (GWWD), 40% of the Nebula watershed is used for annual agriculture in a no-tillage system for soybeans, corn, beans, wheat, barley and oats; 22% of the area is used for pasture and livestock farming for pigs, poultry and cattle; 25% of the region is occupied by forest, and 13% consists of riparian forest. From this scenario, the permanent preservation areas are compromised since most of these activities do not comply with environmental legislation. Another recurring problem concerns washing chemical fertilizer bags and containers in streams that flow into the reservoir, causing eutrophication. Clandestine mining is also known in the region, with the extraction of minerals used in civil construction such as sand, crushed stone, gravel and sand, contributing to erosion and silting up of riverbeds and the dam".

The next day, upon arriving at his research group's laboratory, Thomas meets his friend, Julio, and tells him about the situation at the Nebula Reservoir.

- Since there is no concern for the environment, the activities around the reservoir represent a danger and may be contaminating fish, water and sediments. - Comments Julio.

- Even the sediments?

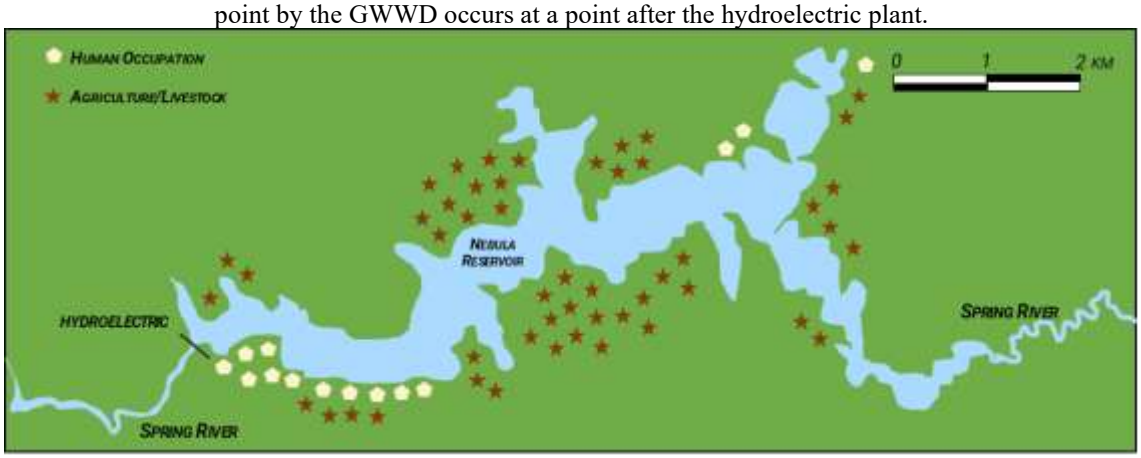
- That's right! Furthermore, metals that are in the water are usually immobilized in sediments. However, physical-chemical alterations or disturbances can make the metals bioavailable again and further contaminate the water and the animals that depend on it.

- Can you help me carry out an analysis of the reservoir sediments then?

- Of course! I can help you.

Thomas shows Julio the map he found of the Nebula region. Together, they begin to think about the sediment sampling points that will support the investigation they intend to conduct. In addition, they speculate on how to analyze the sediments.

Figure 2. Map of the Nebula Reservoir region. Greenfields is located to the left of the map, and the water collection point by the GWWD occurs at a point after the hydroelectric plant.



Activities – Part II

1. Considering the problem proposed by your group in Part I, demarcate points for the collection of sediment samples to help Thomas and Julio in their investigation. Justify your choice of sampling points.

2. Helping Thomas and Julio analyse the sediments is also a mission of your group. Search for two instrumental analytical techniques that can determine the concentration of heavy metals (Al, Cd, Cr, Cu, Fe, Mn, Ni, Pb, Zn) in **sediment samples and argue in favour of the one considered the most appropriate by your group**. The argument can be based on the following aspects: cost of equipment and analysis, complexity of manipulation, sample throughput, detection limits, sample size, and other factors the group find interesting.

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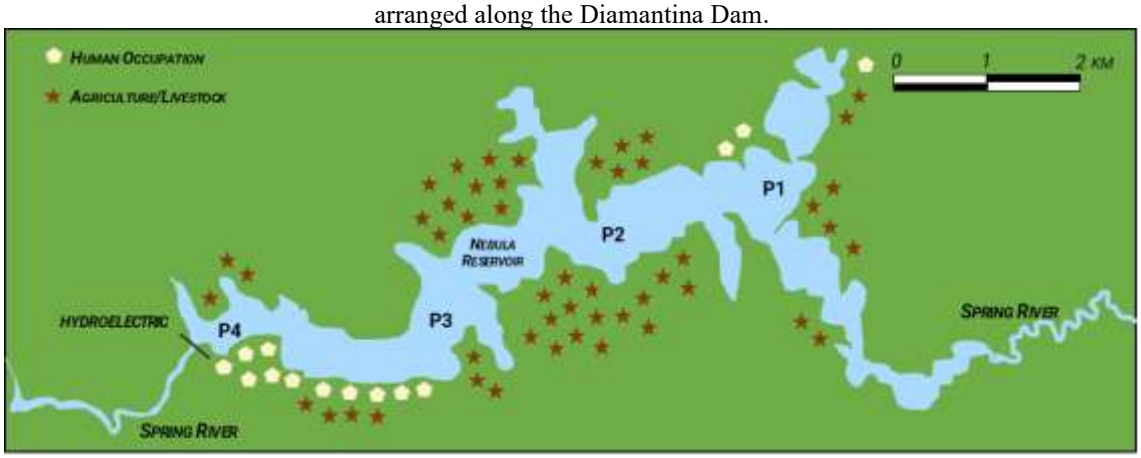
Part III – Diving Deep into the Nebula Reservoir

Thomas and Julio chose four sediment collection points from the previously presented map. Figure 3 shows the location map of the reservoir and the chosen sampling points.

Figura 3. Study area location map. Thomas and Julio demarcated the four sampling points identified on the map arranged along the Diamantina Dam.



Thomas and Júlio went to Diamantina to collect sediment samples the following day. For this procedure, they use an Ekman–Birge collector, performing the collection at the deepest level of the dam at each collection point in Figure 3. Upon returning to the laboratory, the samples were lyophilized, macerated until homogenized and sieved through a mesh of 53 µm. Then, the levels of pseudo-total and bioavailable metals were determined according to the following experimental procedures:

Preparation of sediment samples for the determination of pseudo-total metals: 0.5 g of each homogenized sample were digested in a digester block at 160 ºC, with the addition of 10 ml of 1:1 HNO_3 at reflux for 10 minutes, followed by the addition of 5 ml of concentrated HNO_3 p.a. at reflux for 30 minutes. The process was repeated until complete digestion and volume reduction of 5 ml. After cooling, 2 ml of 30% H_2O_2 were added, repeating the addition until there were no more bubbles. Finally, 10 ml of concentrated HCl p.a. at reflux for 15 minutes and completing the volume with 50 ml. The undigested part was removed by filtration.

Preparation of sediment samples for determination of bioavailable metals: 1.0 g of each homogenized sample was added to an Erlenmeyer flask, and 25 ml of HCl 0.1 mol/L were added and maintained for 2 hours in a horizontal shaker at 200 rpm before filtering the suspension.

The samples were analyzed for Ag, Al, Cd, Co, Cr, Cu, Fe, Mn, Ni, Pb and Zn using Flame Atomic Absorption Spectrometry (FAAS).

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Activities – Part III

1. a) What connections can be established among the sampling criteria adopted by Thomas and Julio and those your group chose? b) What criteria were used by the characters that were not used by your group and vice versa? c) Do you think it is relevant to change the sampling points proposed by your group? **Justify your answers.**

2. Do you think the equipment proposed by your group should be changed in accordance with what was suggested by the characters? **Justify your answer.**

3. The metal concentration in sediments is commonly determined by **pseudototal**, **bioavailable** and **exchangeable fractions**. Define and exemplify the use of such concepts.

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Part IV – Uncovering the Sediments of the Nebula Reservoir

Doing some research, Julio finds the following information, which he decides to share with his friend, Thomas.

"*Zinc is considered an essential micronutrient and plays an important role in the metabolism of many organisms. However, in high concentrations, the metal can cause serious adverse effects on human health and the biotic environment, especially plants and soil macrofauna. In the aquatic environment, zinc predominantly attaches to suspended material before accumulating in the sediment. On the other hand, cadmium is a low-abundance element in the earth's crust and has harmful effects on organisms. In addition to being present in the sediments, the metal can remain in the interstitial water, be associated with the solid fraction or be redistributed within the sediments. It must be made clear that changes in environmental conditions can affect the bioavailability of these metals so that they can be made available again to the water column, thanks to redox reactions or resuspension processes of physical origin (current), biological (activity of organisms living in sediments) and human (navigation)*"

- That is, the dam situation may be much more complicated than we imagine. By the way, do you remember that we found some eutrophic places? Thomas asks.

- I do remember! Eutrophication occurs mainly due to increased nutrients, especially phosphorus and nitrogen. Right?

- That's right, which then causes the excessive proliferation of organisms such as algae and cyanobacteria. These organisms consume a large amount of oxygen in the dam water so that living beings end up dying.

- What a devastating situation!

- But not everything is lost. With the analyzes we carry out, in addition to determining the bioavailability of metals in the environment, we will also be able to assess the quality and potential for toxicity so that we will be able to predict the risks of these metals for the region's population and the environment. And consequently, provide for remedial actions.

Below are the results found by Thomas and Julio from the experiments carried out according to the procedures indicated in Part III of the case study.

Table 1. Concentration of pseudo total metals in sediments from the Nebula Reservoir at the four collection points.

Sampling Points	Concentration (µg/g)										
	Ag	Al*	Cd	Co	Cr	Cu	Fe*	Mn	Ni	Pb	Zn
P1	4,08 ± 0,32	76,39 ± 2,06	2,14 ± 0,07	27,03 ± 1,53	33,48 ± 0,90	29,20 ± 0,67	44,57 ± 2,31	340,85 ± 9,99	27,69 ± 1,21	27,51 ± 1,38	81,13 ± 2,30
P2	4,55 ± 0,11	72,11 ± 6,52	2,39 ± 0,23	27,34 ± 0,70	39,24 ± 0,73	43,77 ± 0,57	43,59 ± 3,75	277,95 ± 3,38	27,80 ± 1,93	33,49 ± 0,96	71,44 ± 2,50
P3	5,66 ± 0,16	79,01 ± 1,66	2,38 ± 0,26	28,21 ± 0,93	43,33 ± 2,06	44,87 ± 1,52	62,05 ± 6,65	318,50 ± 0,38	24,21 ± 1,42	34,90 ± 1,14	58,51 ± 1,80
P4	5,17 ± 0,32	74,23 ± 2,08	4,07 ± 0,31	29,43 ± 0,69	44,66 ± 2,51	43,46 ± 1,03	62,09 ± 1,90	408,27 ± 7,81	30,88 ± 2,77	31,96 ± 0,55	52,62 ± 1,57
Overall Average	4,86 ± 0,69	75,44 ± 2,96	2,74 ± 0,89	28,00 ± 1,07	40,18 ± 5,02	40,32 ± 7,44	53,08 ± 1,04	336,39 ± 54,53	27,65 ± 2,72	31,96 ± 3,20	65,93 ± 12,82

(*) Results expressed in mg/g, other sediment results expressed in µg/g.

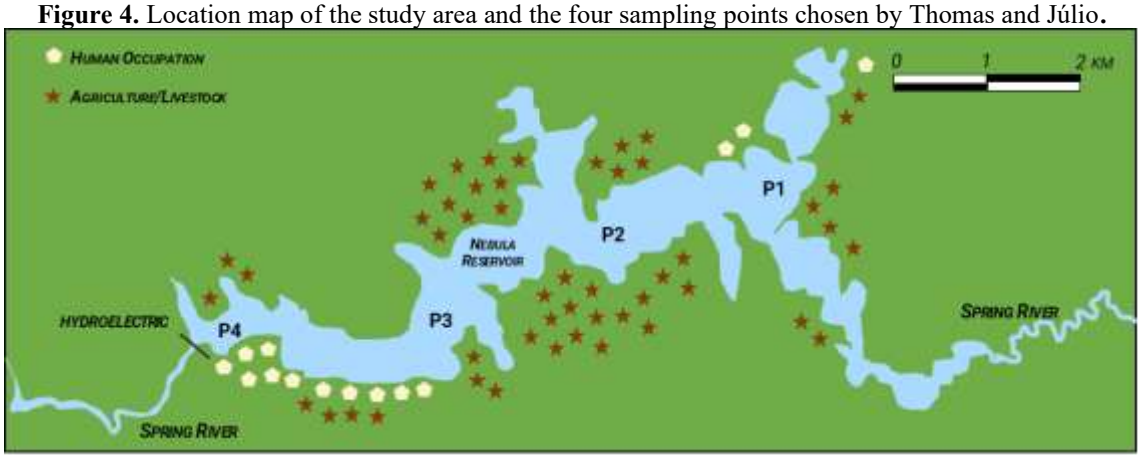
Table 2. Concentration of bioavailable metals in sediments from the Nebula Reservoir at the four collection points.

Sampling Points	Concentration (µg/g)										
	Ag	Al*	Cd	Co	Cr	Cu	Fe*	Mn	Ni	Pb	Zn
P1	0,48 ± 0,06	0,32 ± 0,01	0,03 ± 0,002	1,22 ± 0,22	2,46 ± 0,16	6,12 ± 0,48	3,69 ± 0,30	186,05 ± 6,55	0,84 ± 0,08	2,36 ± 0,17	12,84 ± 0,74
P2	0,77 ± 0,12	0,87 ± 0,03	0,02 ± 0,006	0,33 ± 0,06	2,82 ± 0,20	8,61 ± 0,46	3,71 ± 0,17	154,36 ± 4,21	0,83 ± 0,07	3,08 ± 0,17	7,94 ± 0,37
P3	0,73 ± 0,09	1,11 ± 0,07	0,03 ± 0,004	0,34 ± 0,19	2,83 ± 0,31	8,86 ± 0,42	6,7 ± 0,68	193,27 ± 8,00	0,83 ± 0,11	2,33 ± 0,16	6,65 ± 0,16
P4	0,32 ± 0,02	0,70 ± 0,02	0,007 ± 0,002	0,26 ± 0,06	2,50 ± 0,24	7,15 ± 0,02	6,43 ± 0,51	209,65 ± 11,87	0,76 ± 0,04	1,86 ± 0,07	5,19 ± 0,17
Overall Average	0,57 ± 0,21	0,75 ± 0,33	0,02 ± 0,01	0,54 ± 0,45	2,65 ± 0,20	7,68 ± 1,28	5,15 ± 1,67	185,83 ± 23,19	0,81 ± 0,03	2,40 ± 0,50	8,15 ± 3,32

(*) Results expressed in mg/g, other sediment results expressed in µg/g.

To evaluate the results, Thomas and Julio return to the choice of sampling points (Figure 4). The justification they offered for that choice was the need to contemplate the entire extension of the Diamantina Dam and its level of impact. In this perspective, point one (P1) represents the top of the reservoir; point two (P2) is the area with intensive agriculture; point three (P3) is the beginning of the dwellings; and point four (P4) is close to the hydroelectric plant.

Figure 4. Location map of the study area and the four sampling points chosen by Thomas and Júlio.



Below are the reference values of the Brazilian National Council for the Environment (CONAMA),[1] used by the boys to assess the concentration of metals analyzed in the sediments. Level 1 values represent the threshold level of possible adverse effects on the biological community. That is, they are the maximum values for which the effects of contamination are minimal. Level 2 values represent the threshold level of likely adverse effects on the biological community. That is, concentrations above this level indicate a high possibility of adverse effects. To compare values, it is essential to emphasize that the unit of measurement µg g^{-1} is analogous to mg kg^{-1}; that is, comparing most of the values present in Tables 1 and 2 with the values in Table 3 can be made without the need for conversion calculations.

Table 3. Reference values of maximum permissible concentrations (mg kg^{-1}) for the metals analyzed by Thomas and Julio in the sediments collected at the Diamantina Dam. (-): there is no criterion for this parameter.

Maximum Concentration	Ag	Al	Cd	Co	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Level 1	-	-	0,60	-	37,30	35,70	-	-	18,00	35,00	123,00
Level 2	-	-	3,50	-	90,00	197,00	-	-	35,90	91,30	315,00

[1] BRASIL. Conselho Nacional do Meio Ambiente. **Resolução CONAMA nº 454 de 01 de novembro de 2012.** Estabelece as diretrizes gerais e os procedimentos referenciais para o gerenciamento do material a ser dragado em águas sob jurisdição nacional. Diário Oficial da União, Brasília, 01 de jan., seção 1, p. 66.

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Activities – Part IV

1. What connections can be established between the data acquired by Thomas and Julio (Tables 1 and 2) and the sampling points they chose (Figure 4) for the understanding the influence of anthropic activities in the region on the aquatic life of the Nebula Reservoir?

2. What conclusions can be drawn on the adverse effects on the environment in the Nebula Reservoir region from the comparison of the data obtained by Thomas and Julio (Tables 1 and 2) with reference values for metals in sediments (Table 3)? **Justify your answer.**

3. Did the data interpreted so far confirm the hypotheses formulated by your group in Part I of the case study? **Justify your answer.**