

Microbiological quality of water from the Corredor Stream in Mario Campos, Minas Gerais, in the metropolitan region of Belo Horizonte

Qualidade microbiológica da água do Córrego Corredor em Mário Campos, Minas Gerais, região metropolitana de Belo Horizonte

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ABSTRACT

In 2019, a sewage treatment station was installed by the Municipality of Mario Campos on the banks of the Corredor Stream in the Bela Vista neighborhood, which releases this treated sewage directly into the stream. This altered the water's colorimetric aspect and induced a strong odor, inconveniencing the local population who, in addition to suffering from the bad smell, are unaware of the quality of the water in question. Even so, this water is still used to irrigate part of the leafy vegetables which feed the local population and a large part of the Belo Horizonte metropolitan region. In 2021, the present study began with the aim of evaluating the microbiological parameters of the stream water before and after the discharge of treated sewage and comparing them to the potability standards described in CONAMA Ordinance no. 357/2005. Analyses for the presence of coliforms were conducted according to the FUNASA Manual (Brazil, 2006), and by analyzing a multiparametric probe to determine physicochemical patterns. The presence of bacteriophages was also evaluated, using *Escherichia coli* as bait. Water conditions were far from desirable based on microbiological parameters, often exceeding the limit of 100 CFU/mL, indicating excessive levels of total and thermotolerant coliforms. Coliphages were detected, another bioindicator that confirms the presence of *E. coli*. The physicochemical standards also did not meet those required for potability. This is a pioneering study in investigating the water quality of the Corredor Stream and shows that it is unsuitable for the irrigation of vegetables eaten raw.

Keywords: Coliforms. *Escherichia coli*. Water analysis.

RESUMO

Em 2019, foi instalada pela Prefeitura de Mário Campos, às margens do Córrego Corredor, no bairro Bela Vista, uma estação de tratamento de esgoto que lança o esgoto tratado diretamente no córrego. Esse fato alterou o aspecto colorimétrico e induziu um forte odor, causando transtornos à população local, que, além de sofrer com o mau cheiro, desconhece a qualidade da água em questão. Mesmo assim, a população utiliza essa água para irrigar parte das hortaliças folhosas que alimentam a população local e grande parte da região metropolitana de Belo Horizonte. Em 2021, o presente trabalho teve início com o objetivo de avaliar os parâmetros microbiológicos da água do córrego antes e depois do lançamento do esgoto tratado e compará-los aos padrões de potabilidade descritos na Resolução Conama n.º 357/2005. As análises de presença de coliformes foram realizadas conforme o Manual da Funasa (Brasil, 2006) e por análises com sonda multiparamétrica para determinação dos padrões físico-químicos. A presença de bacteriófagos foi avaliada utilizando *Escherichia coli* como isca. As condições da água estão distantes do desejável com base nos parâmetros microbiológicos, ultrapassando muitas vezes o limite de 100 UFC/mL, indicando níveis excessivos de coliformes totais e termotolerantes. Foi detectada a presença de colifagos, bioindicador que confirma a presença de *E. coli*. Os padrões físico-químicos também estavam em desacordo com os exigidos para potabilidade. Este estudo é pioneiro em investigar a qualidade da água do Córrego Corredor e evidencia que a água está imprópria para a irrigação das hortaliças consumidas cruas.

Palavras-chave: Análise de água. Coliformes. *Escherichia coli*.

INTRODUCTION

Water is a natural resource that plays a central role in various cycles and phenomena that ensure the ecological maintenance of ecosystems and life on the planet. However, this essential resource is being threatened by uncontrolled urbanization, which causes changes in the hydrological cycle, such as soil impermeability, river channeling, water pollution and the reduction of biodiversity. Water is used in various activities, whether in agriculture, industry or for consumption, and therefore, knowing the potability standards matters greatly for health and its appropriate usage (Tucci, 2008).

However, approximately 35 million Brazilians lack access to treated water, and it is estimated that approximately 100 million inhabitants have no sewage treatment in Brazil, resulting in diseases which may lead to death (SNIS, 2018). This critical situation is directly linked to the increase in infectious diseases, especially diarrheal diseases. Studies published in the literature that evaluate the results of sanitation interventions have indicated a significant positive effect on intestinal infectious diseases, which resulted from improvements in access to water and sanitation. The evidence on the magnitude of the effect of unimproved access to water and sanitation on the relative risk of

diarrhea is clear, showing the urgency of effective interventions in this sector (Rêgo, Killinger & Barreto, 2018).

This can be corroborated by the statements of the Trata Brasil Institute (2023), which reports that almost 200,000 Brazilians were hospitalized due to waterborne diseases (Abcon Sindicon, 2023). It is also suggested that these figures may have been higher, since, due to the pandemic, hospital beds used to prioritize patients with Covid-19. This observation emphasizes the importance of basic sanitation as a preventive measure against infectious diseases.

Worldwide, waterborne diseases are the leading cause of death in childhood, with diarrhea being one of the main aggravating factors, caused by several groups of bacteria, viruses and parasites. Examples of these infections include cholera, salmonellosis, shigellosis, amoebiasis, balantidiasis, giardiasis, cryptosporidiosis, isosporiasis, rotavirus enteritis, acute gastroenteropathy caused by Norwalk agent, adenovirus enteritis, other types of viral enteritis, enteric fevers, typhoid fever and hepatitis A (Costa et al., 2002). These diseases also compromise the work capacity and overload capacity of healthcare systems. Sanitation services must prevent the population, water supplies,

disease vectors and food from encountering human waste. However, the absence of this type of service is observed in the vicinity of the Corredor Stream in the Municipality of Mario Campos, which increases concern about diseases related to inadequate sewage treatment.

Though it belongs to the metropolitan region of Belo Horizonte, the Municipality of Mario Campos has characteristics of rural areas, with the use of water from wells, cisterns, streams, rivers and lakes for economic development and leisure (Ramos, Silva, Lima, Marques & Gontijo, 2020). It is already a recognized fact that investments in sanitation and water quality prevent diseases and improve the quality of life of the population. In this scenario, microbiological control measures are suggested for the prevention of diseases and analysis of contaminated samples (Fernandes et al., 2018).

Mario Campos is one of the largest producers of vegetables in the state of Minas Gerais, responsible for the supplying of the capital of Minas Gerais and the entire metropolitan region, as well as other large municipalities, including Sete Lagoas (Rodrigues & Tubaldini, 2002). In this sense, vegetable production is of great significance to the local economy (Mariano, 2023) because in addition to generating income, it supplies consumers in downtown Belo Horizonte and cities such as Ouro Preto, Sete Lagoas and the surrounding regions of these municipalities, through CEASA5, reaching regions outside the state of Minas Gerais.

Moraes, Costa e Silva, Bastos and Souza (2017) mapped the land use and occupation of Mario Campos and found that over the years there has been a decrease in areas covered by dense vegetation and an increase in areas occupied by agricultural crops throughout the municipal territory. This highlights how relevant their local agricultural activity is. And despite its importance in creating jobs and as a source of income for the municipality, there are no studies on the quality of the water used for such activity, especially for irrigating the vegetables produced.

The Corredor Stream is located in Mario Campos, Minas Gerais, and is part of the Paraopeba River basin. In 2019, a sewage treatment plant (STP) was built to collect domestic sewage from two nearby neighborhoods, Bela Vista and Tangará, with approximately 400 homes (according to data provided by the city government). However, this STP has only acted as a collector, so the effluent released into the body of water does not have the visual and sensory traits compatible with the proposed treatment and the sewage does not present a clear and chlorinated aspect at the time of disposal. This situation has caused discomfort for the surrounding population and altered the water quality downstream from the effluent reception site.

According to Von Sperling (2005), after secondary treatment to remove dissolved and suspended organic matter, sewage already has characteristics that allow it to be released into the environment. However, in the aforementioned case, a large amount of organic matter is still visible at the time of release. According to reports from residents, this stream was used not only for irrigating vegetables, but also for recreation and fishing. Ever since the introduction of the STP, the water has become cloudy and smelly, but farmers still use pumped water to irrigate vegetables that are eaten raw or peeled. Since there are no records of water analyses for the Corredor Stream in the literature, this study proposed the analysis that was carried out throughout 2021 to verify the quality of the water in question used for irrigating vegetables eaten raw and to assess the effectiveness of the sewage treatment plant on site.

Physicochemical analyses are important parameters for inferring water quality. Such analyses are essential in the food industry, for example, as this factor can indicate the presence of

residues generated by sanitation, microbiological contamination or even cross-contamination. Therefore, measuring the physicochemical parameters of water can help and serve as an indicator of water quality. Checking turbidity, potential of hydrogen and the amount of dissolved oxygen in the water and identifying and monitoring the presence of harmful substances, such as chemical pollutants and organic solids, makes it possible to draw inferences about the survival of pathogens which may pose health threats. Therefore, investing in water monitoring and analysis not only prevents health risks, but also contributes to environmental preservation, ensuring access to safe, quality water, which is essential for the well-being and quality of life of the population (Fernandes et al., 2018).

Microorganisms present in water can be used as bioindicators of water quality and of the degree of impact on the ecosystem. The main organisms found in sewage are bacteria, fungi, protozoa, viruses and algae. Among these, bacteria are the most used as indicators since they are responsible for the decomposition and stabilization of organic matter. A specific group, thermotolerant coliforms, are the most used bioindicators because they serve as proof for the contamination of water with feces from homeothermic animals, since they inhabit the intestinal tract of these animals (Fernandes et al., 2018). Therefore, the 2005 CONAMA Resolution and the Consolidation Ordinance no. 05/2017, by the Ministry of Health (Brazil, 2008), consider microorganisms associated with the intestinal microbiota as bioindicators of water quality because they show contamination by feces (Seta, Oliveira & Pepe, 2017).

Another indicator that favors the confirmation of fecal contamination is the presence of coliphages that are specific to *Escherichia coli*, an intestinal bacterium. The presence of such organisms not only confirms the presence of these bacteria in the water but is also an indirect indication that human enteric viruses are present. Due to the physical and chemical characteristics of viral particles, they can remain in the water even after bacterial elimination and may be associated with outbreaks of diarrhea of viral origin. Due to their abundance and evolutionary traits, they can be used as a biological tool for the treatment of infectious diseases, as well as to identify and/or remove bacteria from the environment (Apolinário, 2023).

The Paraopeba River basin supplies water for an estimated population of 180,204 inhabitants (Brazilian Institute of Geography and Statistics [IBGE], 2019). Some of its tributaries had their basins protected by the legislation of the state of Minas Gerais in the mid-1980s and early 1990s, and some of its tributaries (Balsamo, Rola-moça, and Taboões streams) had their basins protected by state legislation in order to guarantee the supply of drinking water to part of the metropolitan region of Belo Horizonte (Alves, Maia, Lazaro, Cota & Magalhães, 2022). The Corredor Stream, located in Mario Campos, is part of the Ribeirão Sarzedo sub-basin, another tributary of the Paraopeba River.

Due to the lack of information on the water quality of the Corredor Stream, the population of this region continues to use its water. Many are unaware of the existence of the quality indicators required for the use of this water in vegetables that are eaten raw and sold, with the risk of being a source of human contamination. In this sense, the aim of this study was to verify the quality of the water from the Corredor Stream that is used for irrigating vegetables in the municipality before and after the waste discharged by the new sewage treatment plant (STP) introduced near this stream. It is believed that its environmental impact may have increased the water's microbial population, especially the presence of the bioindicator *E. coli*.

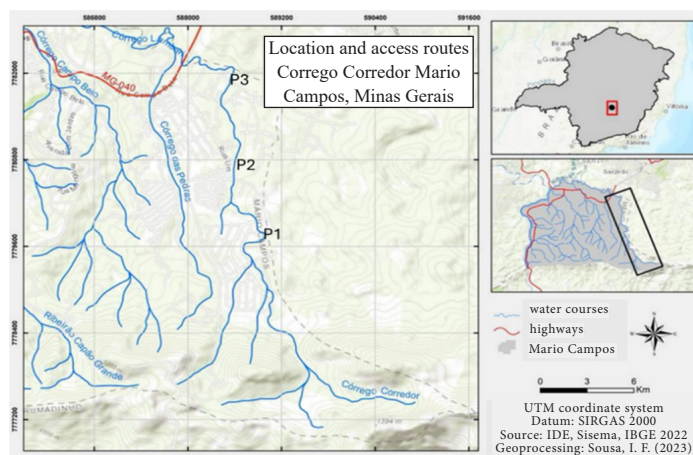
MATERIALS AND METHODS

The city of Mario Campos has an area of 72,395 km². Geomorphologically, the municipality is in the Metallurgical Zone, being part of microregion no. 182 (Belo Horizonte). The study was conducted in the Corredor Stream, located in this municipality and belonging to the Ribeirão Sarzedo sub-basin and the Paraopeba River basin. Three sampling points were defined and chosen to assess whether the sewage treatment plant (STP) installed on site has maintained the analyzed parameters similar to those observed at the point that precedes the installation of this STP: P1 – located in the region close to the source of the stream (20° 4'28.32" S; 44° 9'5.43" W); P2 – represents the site after the sewage discharge from the STP (20° 4'13.16" S; 44° 9'8.44" W); P3 – located downstream from the effluent discharge treated by the STP and close to the vegetable gardens that use water from the stream (20° 3'19.79" S; 44° 9'21.18" W). These points were chosen to delimit an interval from a point before the STP (P1), to a point right after the discharge of sewage treated by the STP (P2) and another point after the STP that is next to 1 kilometer from the STP (P3) in order to verify whether the treated sewage has influenced the presence of thermotolerant coliforms.

The Mario Campos watersheds, indicating the stream studied, can be visualized in Figure 1, obtained from the State System of Environment and Water Resources (SISEMA, 2022).

Figure 1

Map of the river basins of the Municipality of Mario Campos, Minas Gerais, indicating the Corredor Stream. P1 – first sampling point, after the spring; P2 – second sampling point, after the STP; P3 – third sampling point, near the vegetable gardens.



Source: SISEMA, 2022.

Sample collection

Samples were always collected in the morning between 7:30 a.m. and 8:00 a.m. during the months of February, March, May and July 2021. The intervals were due to rainy periods which affected the availability of materials to perform the analyses and in which sampling could not be done. Water was collected directly from the subsurface of the water body (0 to 30 cm deep) in a sterilized, wide-mouth glass container with a well-fitted, ground glass lid, with a maximum capacity of 125 mL. Samples for microbiological analyses were collected in triplicate and kept between four and 10 °C during shipping to the laboratory, according to the methods specified by the Manual proposed by FUNASA (Brazil, 2006). Analyses always began within one hour after collection. The samples for phytoplankton analysis were collected in an amber glass bottle, fixed with acetic lugol and stored in a dry and ventilated place until the time of counting.

Collection of physical and chemical data

During sampling, water temperature, electrical conductivity, pH and turbidity parameters were analyzed on site, using a Horiba U-50 multiparameter probe.

Microbiological analyses

Analyses of total and thermotolerant coliforms, as well as the presence of bacteriophages, were performed at the UEMG Applied Microbiology Laboratory (State University of Minas Gerais). All procedures were performed in a laminar flow hood under sterile conditions. Bacteriological tests were performed using the multiple tube technique, which consists of demonstrating the quantity of coliforms in the Most Probable Number (MPN) per 100 mL⁻¹ sample. The total coliform test is subdivided into two tests, namely the Presumptive Test and the Confirmative Test, according to the FUNASA Manual (Brazil, 2006).

Total coliforms

For the presumptive test, 15 test tubes were used per sample point, five of which contained double-concentration lactose broth and ten contained single-concentration lactose broth. In the first five tubes (double concentration), 10 mL of the collected water sample were inoculated (dilution was 1:1). In the remaining ten tubes (single concentration), 1 mL of the sample was inoculated, in the first five with a dilution of 1:10. In the last five tubes with a dilution of 1:100, 0.1 mL of the sample was inoculated. All tubes were incubated at 35 °C ± 0.5 °C for 24/48 hours. After this period, gas formation inside the Durham tube and turbidity of the medium were verified, confirming that the presumptive test was positive. Subsequently, the confirmatory test was performed and with the previously flamed and cold subculture loop, a portion of the sample was removed from each positive tube and inoculated into the appropriate tube containing 2% (v/v) Brilliant Green Bile medium. The tubes were incubated for 24/48 hours at 35 °C ± 0.5 °C and the presence of gas inside the Durham tube confirmed the test as positive. Results were expressed in M.P.N. (Most Probable Number) x 100 mL⁻¹ of sample. To determine the M.P.N., the combination formed by the number of positive tubes that presented in the dilutions 1:1; 1:10; 1:100 in the Confirmatory Test was verified.

Thermotolerant coliforms

To identify thermotolerant coliforms, the positive presumptive test tubes (gas formation) and all negative tubes in which there was growth after 48 hours, in the three dilutions (1:1; 1:10 and 1:100), were analyzed. A portion collected with a flamed and cold subculture loop was inoculated into the corresponding test tubes containing the EC medium, prepared according to the manufacturer's recommendations. The tubes were incubated in a water bath at 44.5 °C ± 0.2 °C for 24 ± 2 hours. After 24 hours, the formation of gas inside the Durham tubes confirmed the presence of thermotolerant coliforms. After this verification, the M.P.N. was calculated based on the table provided by the National Health Foundation's Practical Manual for Water Analysis (Brazil, 2006).

Total bacterial count

Plate Counter agar prepared according to the manufacturer's recommendations was used for the total bacteria count. Using a volumetric pipette, 1 mL of the collected water sample was transferred to a previously sterilized Petri dish. Then, the culture medium previously melted and stabilized in a water bath at 44 °C-46 °C was added to the water deposited on the plate.

The contents of the plate were homogenized in moderate circular movements and the plate was incubated at $35\text{ }^{\circ}\text{C} \pm 0.5\text{ }^{\circ}\text{C}$ for 48 ± 3 hours. At the end of the incubation period, colonies were counted and results were expressed as the number of bacterial colonies per milliliter or Colony Forming Units (CFU) per milliliter.

Confirmation of the presence of *E. coli* in selective and differential medium

To verify the presence of *E. coli*, Methylene Blue Eosin agar was also used. This agar is selective and differential for enterobacteria and allows for the growth of *E. coli* in a bright green color.

Analysis of the presence of *E. coli* using coliphages

For this analysis, the collected water samples were centrifuged for 30 minutes at 3000 g to remove solid debris and filtered with a $0.22\text{ }\mu\text{m}$ pore-sized polyethersulfone (PES) filter membrane. One milliliter of the filtrate was added to 10 mL of *E. coli* 30 bacteria culture, used as bait, with 10^8 CFU/mL. This culture was incubated for 24 hours and then centrifuged to remove the bacterial suspension, and the supernatant was filtered using a $0.22\text{ }\mu\text{m}$ pore-sized membrane (Kutter, 2009).

Isolation was performed by observing the formation of lysis plaques on an agar layer according to the methodology adapted from Kęsik-Szeloch et al. (2013). For this purpose, 250 μL of *E. coli* M30 culture (1×10^6 cells) previously incubated at $35\text{ }^{\circ}\text{C}$ to reach the exponential growth phase, approximately four to five hours, were mixed with 50-100 μL of viral filtrate and the mixture was added to 5 mL of Luria Bertani medium containing 0.75% agar, previously maintained at $55\text{ }^{\circ}\text{C}$. The mixture was poured onto plates containing a thin layer of solid LB medium containing 1.5% agar. The plates were incubated overnight, and lysis plaques were observed. All procedures were performed in triplicate from samples that were collected in March, May and July, after high coliform concentrations were observed in the previous months.

Phytoplankton Counts

Phytoplankton counts were performed under an inverted microscope, using the Utermöhl sedimentation technique (Utermöhl, 1958). One hundred individuals of the dominant species were counted, respecting the 80% confidence limit (Lund, Kipling & Le Cren, 1958). Each unicellular, colonial and filamentous alga was considered an individual.

RESULTS AND DISCUSSION

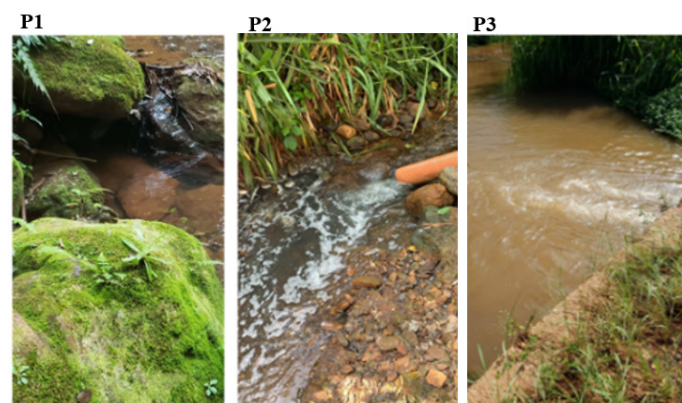
The physical-chemical analyses were performed at the sampling site, while the microbiological analyses were performed on water collected at points P1, P2 and P3, the photos of which can be seen in Figure 2. The analyses performed at this stream are the first to be released, and there is no information on the quality of this water before the STP was installed. The data for the water at point P1, before the location where the STP was built, are within the parameters established by CONAMA in its resolution that addresses the presence of thermotolerant coliforms in water used for irrigation of vegetables eaten raw. The number of countable bacteria was above 1600 CFU/mL at points P2 and P3 in all samples, while P1 remained at 200 CFU/mL. At points P2 and P3, the sampled parameters show that such water is unusable for this purpose. The visual aspect of the images presented in Figure 2 already allows us to observe the eutrophication of the water when comparing points P1, P2 and P3.

With the exception of point P1, the sampled water presented a coliform concentration that was well above the

desirable levels for its intended use, namely the irrigation of vegetables (Figure 3), since, according to CONAMA Resolution no. 357, the maximum limit of coliforms present in water intended for the irrigation of vegetables eaten raw must not exceed 200 MPN and, in most cases, the limit found for points P2 and P3 was above 1600 MPN, hence disqualifying the water for this purpose. In Brazil, only 46% of the country's sewage receives treatment (SNIS, 2018) and this situation is worrying, since, according to studies, many watercourses are used for consumption, irrigation and other purposes.

Figure 2

Sampling collection sites for the microbiological and physical-chemical tests performed in this study. P1 – Near the spring next to Serra dos Três Irmãos; P2 – After the sewage discharge from the existing STP; P3 – Near highway MG 040.



Source: The authors.

Based on the data shown in Table 1, one observes that the amount of *E. coli* presented in points P2 and P3 is above the desirable level, with average values above 500 MPN. This result is in line with Santos (2007), who points to a direct relationship between the increase in economic activities of the urban population and the large scale of impurities in the waters of streams, rivers and lakes. The data also agree with the scenario that Alves et al. (2022) highlight in their studies, in which 80% of domestic and industrial effluents in Brazil are discharged directly into springs, rivers and lakes without any prior treatment.

Such data proves unsatisfactory in light of the CONAMA Resolution no. 357/2005 when it sets the criteria for water to be used in agriculture for the irrigation of vegetables that can be eaten raw or fruits that grow close to the ground and are eaten raw without having their coating removed. In fact, these data confirm the overview on water supply and sanitation presented by the State Secretariat for the Environment and Sustainable Development in 2020 (SEMAD, 2020). The aforementioned report indicated that, in the Municipality of Mario Campos, only 14% of the population has access to sewage collection, out of which only 9.9% have access to sewage treatment. This picture classifies the city as being in a critical state, leading to the conclusion that it must develop public policies and initiatives for the improvement and creation of sewage treatment plants (STPs).

Our data, similarly to the findings of a study by Rocha et al. (2006) conducted in the Santa Cruz Basin at the city of Lavras, state of Minas Gerais, revealed that the samples were contaminated with thermotolerant coliforms and that the water did not meet the mandatory requirements. This sampled water is used to irrigate vegetables close to the ground or meant to be eaten raw. The aforementioned resolution defines this type of water as Class I due to its purpose, and yet, the water used was found to be well below the expected standards.

When analyzing the Eosin Methylene Blue medium plates inoculated with the samples from the positive tubes of Brilliant Green Bile Broth, the presence of metallic green colonies is noted, as shown in Figure 3A. This allows macroscopic confirmation of the presence of thermotolerant *E. coli* in the samples analyzed. In addition to confirmation from plates, the Gram technique was performed and, through this, bacterial cells in the form of bacilli with a pink coloration were visualized, confirming the presence of Gram-negative bacteria (Figure 3B), thus showing that this is the microorganism indicating fecal contamination by *E. coli*.

Table 1

Most Probable Number (MPN) of total and thermotolerant coliforms from February to July 2021, according to series five of multiple tubes.

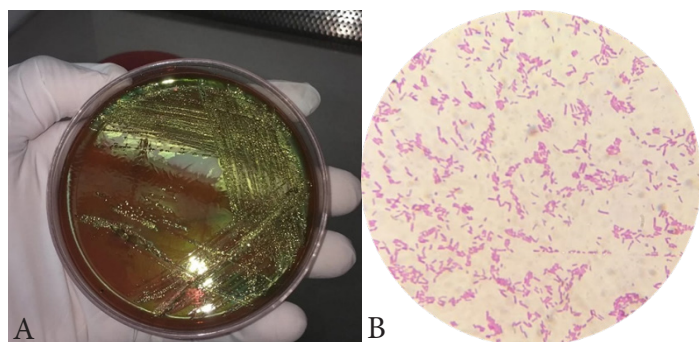
| Month | Sampling points | Microbiological | |
|----------|-----------------|-----------------------------|--------------------------------------|
| | | Total Coliforms *MPN/100 mL | Thermotolerant Coliforms *MPN/100 mL |
| February | P1 | 350 | 600 |
| | P2 | 900 | 900 |
| | P3 | 1600 | 900 |
| March | P1 | 350 | 350 |
| | P2 | ≥ 1600 | 1600 |
| | P3 | ≥ 1600 | 1600 |
| May | P1 | 500 | 500 |
| | P2 | > 1600 | 1600 |
| | P3 | ≥ 1600 | 1600 |
| July | P1 | 500 | 240 |
| | P2 | ≥ 1600 | 380 |
| | P3 | ≥ 1600 | 500 |

Source: The authors.

Notes. *MPN with 95% confidence limit for various combinations of positive results when five tubes are used for each dilution (10 mL, 1.0 mL and 0.1 mL), as recommended in the table from the FUNASA Manual (Brazil, 2006).

Figure 3

(A) Macroscopic confirmation of the presence of *E. coli* in Methylene Blue Eosin agar; (B) Confirmation of the presence of *E. coli* in samples analyzed by the Gram technique.



Source: The authors.

Confirmation of the presence of these bacteria in numbers that exceed those stipulated by Brazilian legislation shows that the water sampled at Corredor Stream, from the second collection point (P2) located right after the current STP discharge onwards, is unfit for consumption or irrigation of vegetables. Despite this, during the collections it was observed that some farmers still pump water from this stream to irrigate their vegetable gardens, and most of those vegetables are meant to be eaten in their raw

form, which poses a very serious situation, considering that eating these vegetables without proper hygiene may lead to diseases caused by waterborne microorganisms and parasites.

Initiatives to mitigate these problems are necessary, since a large portion of the municipality's population uses the contaminated waters of the Corredor Stream to plant vegetables, which are shipped throughout the state of Minas Gerais, according to reports available both on the city's website and in academic studies (Pimenta, 2016).

The three collection points presented average pH values within the parameters established in CONAMA Resolution no. 357/05 (Brazil, 2005) for Class I water bodies, which stand between six and nine. Razmi et al. (2023) characterize pH values as a significant environmental factor that controls bacterial growth, their activity and affects their metabolic properties, taking into account that most bacteria have an optimal pH between 6.5 and 7.5. Point P1 had an average pH of 7.55, point P2 had an average pH of 7.4 and point P3 had an average pH of 6.84. Overall, such values found for the pH parameter (Table 2) favor microbial growth and corroborate the microbiological data found in the study.

Table 2

Hydrogen potential (pH) of the water from Corredor Stream verified for points P1, P2 and P3 in the months of February, March, May and July 2021.

| | February | March | May | July |
|----|----------|-------|-----|------|
| P1 | 6.9 | 6.7 | 8.9 | 7.7 |
| P2 | 6.3 | 8.3 | 8.3 | 6.8 |
| P3 | 6.7 | 6.6 | 6.6 | 6.2 |

Source: The authors.

Concerning the variation in electrical conductivity, CONAMA Resolution no. 357/05 (Brazil, 2005) establishes that such variation for natural waters should be between ten and 100 $\mu\text{S}/\text{cm}$, while those containing pollutants are in a margin greater than 1000 $\mu\text{S}/\text{cm}$. On conductivity (Table 3), point P1 had an average of 493 $\mu\text{S}/\text{cm}$, point P2 had an average of 223 $\mu\text{S}/\text{cm}$ and point P3 had an average of 101 $\mu\text{S}/\text{cm}$. According to Santos (2007), the conductivity value and contamination level are directly linked and, therefore, one may argue that the higher the electrical conductivity value sampled in water, the greater the contamination of this water resource will be. In this case, all points presented an average conductivity above the established level. The high values at points P1 and P2 may be related to the accumulation of dissolved ions in the water body, requiring an analysis of the solutes present in the stream water to confirm this hypothesis (Esteves, 2011).

Table 3

Electrical conductivity ($\mu\text{S}/\text{cm}$) of the water from Corredor Stream verified for points P1, P2 and P3 between the months of February, March, May and July 2021.

| Sampling points | February | March | May | July |
|-----------------|----------|-------|-------|-------|
| P1 | 0.680 | 0.065 | 0.600 | 0.063 |
| P2 | 0.177 | 0.204 | 0.251 | 0.261 |
| P3 | 0.084 | 0.104 | 0.097 | 0.120 |

Source: The authors.

Temperature influences several other chemical and physical parameters and is a fundamental environmental factor

for controlling aquatic life. Water temperature is also influenced by other factors, such as altitude and latitude, season of the year, rainfall rate, depth of the water body and the flow of its current (Santos, 2007). When the average water temperature of the stream was assessed (Table 4), at point P1 temperature was measured at 19.37 °C, at point P2, 21.12 °C, and at point P3, 19.87 °C. The data that was found indicated adequate temperatures for all points, as recommended by the CONAMA Resolution. There was some variation in temperature values in the months of sampling, mainly in relation to July compared to the other months, between 1.8 °C and 5.5 °C. The low temperature found in the winter months is therefore due to the decrease in atmospheric temperature, which influenced the average temperature for Corredor Stream at that time, when a sharp drop in the minimum temperature was recorded in the metropolitan region for the year 2021 according to Brazil's National Institute of Meteorology (2021).

Table 4

Water temperature (°C) of the Corredor Stream recorded at points P1, P2 and P3 between March and December 2021.

| | February | March | May | July |
|----|----------|-------|------|------|
| P1 | 20.7 | 19.3 | 16.8 | 20.7 |
| P2 | 22.5 | 21 | 19.2 | 21.8 |
| P3 | 22.4 | 18.7 | 16.6 | 21.8 |

Source: The authors.

Turbidity may be defined as a measure of the degree of interference in the passage of light through the liquid (Estevam, Silva and Silva, 2019). It is considered a physical characteristic of water and occurs due to the presence of substances suspended in a liquid medium, such as: divided solids or in a colloidal state, suspended organic matter, insoluble minerals, inorganic particles and microscopic organisms (Santos, 2007). Furthermore, the presence of these particles causes the dispersion and absorption of light, which gives the water a cloudy aspect, aesthetically undesirable and potentially dangerous, since a high turbidity rate points to the blocking of light, thus impairing the photosynthesis of submerged algae and aquatic plants.

The data presented indicated turbidity values above those recommended at point P2. As established by CONAMA Resolution no. 357/05 (Brazil, 2005), turbidity values must be below 40 NTU, which indicates a change in the physical aspect of the water. Point P2 presented on average values above 40 NTU (Table 5), as it receives sewage that underwent some treatment by the STP built by the Mario Campos city government. This data suggests some irregularity in the quality of the water discharged into the stream by the STP, since the other points presented values that were within the quality standard. At point P2, this high turbidity rate is due to the proliferation of microorganisms in the water that receives treated sewage, since it coincides with the highest number of thermotolerant coliforms sampled at the site.

The physical-chemical data found in our study are similar to those found by Kauano, Valerio & Andreola (2018) at the Moscados Stream in the city of Maringá, Paraná. The authors report that some parameters, such as pH and conductivity through the Horiba probe, are within the maximum values allowed by the CONAMA Resolution no. 357/2005, but despite this, the presence of coliforms and increased water turbidity suggest contamination of the stream in question. Such data are also similar to those found by Lenz, Lieberknecht & Machado (2024), who, studying the physical-chemical parameters of the Fiuza River in the city of Panambi, Rio Grande do Sul, found that along the course of the river there was a point where turbidity was higher than in the

other sampled locations and concluded that this occurred due to the greater accumulation of materials associated with discharges from the nearby urban and industrial areas. In the case of the Corredor Stream, this increase can also be attributed to the STP at point P2.

Table 5

Water turbidity (NTU) of the Corredor Stream verified for points P1, P2 and P3 between the months of March and December 2021.

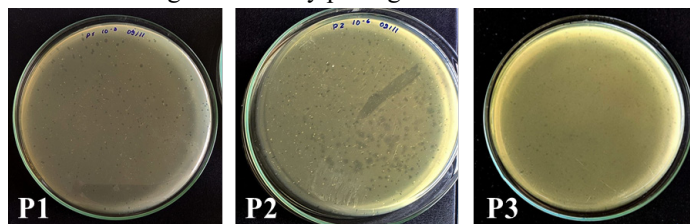
| | February | March | May | July |
|----|----------|-------|-----|------|
| P1 | 4 | 4 | 1 | 5 |
| P2 | 34 | 42 | 51 | 41 |
| P3 | 37 | 20 | 7 | 21 |

Source: The authors.

During the research, we also sought to determine the presence of bacteriophages in the water bodies studied. This strategy was devised because phages are indicators of fecal contamination or inadequate sewage treatment. Bacteriophages are believed to be the most abundant life form on the planet; they can be found in soil, fresh water and salt water, plants and animals, and are ubiquitous (Pereira, 2011). In this study, phages capable of causing the lysis of *E. coli* 30 were detected at all sampling points, as can be seen in Figure 4.

Figure 4

Presence of phages in samples from points P1, P2 and P3, observed through the overlay plating method.



Source: The authors.

At points P2 and P3, the presence of phages was expected due to the assumption of fecal contamination of the water, while at the first point the presence of lysates was unlikely. This detection of phages at P1 may be related to the surface runoff of rainwater, which ends up carrying waste from animals that inhabit farms near the sample point. "Surface runoff from rainwater is considered a source of diffuse pollution and its study can help with sanitation [...]" (Faustino, 2021, p. 8).

The data from the coliphage analysis confirms the presence of *E. coli* in the stream and the characterization of these phages found in the present study will allow us to understand their potential as an agent for analyzing fecal contamination, as well as an agent for controlling infections by permissive bacterial strains. In the first test, these phages were able to infect an *E. coli* strain, and this may be useful for future studies designed for phage therapies, for example. Additionally, the use of bacteria with well-defined protocols will help to establish the level of influence that sewage has on stream contamination and possible diseases or bacterial resistance that may be related to the presence of these phages. They represent one of the mechanisms of bacterial gene variability by transduction and can transfer genes from one bacterium to another, making them, for example, resistant to antibiotics to which they were previously sensitive (Brown-Jaque, Calero-Cáceres & Muniesa, 2015).

Periasamy and Sundaram (2013) demonstrated the effectiveness of using bacteriophages as indicator agents and

growth inhibitors for different pathogenic bacterial strains of the Enterobacteriaceae family found in sewage samples from different hospitals in India. These findings are consistent with the data from the present study, since, in addition to finding phages that multiply specifically in *E. coli*, these microorganisms were isolated in samples of water from a stream used by nearby residents.

In addition to bacteriophages, the search for parasites can help diagnose the quality of water used to irrigate vegetables eaten raw, as was done by Ragazzi (2011). This author found that vegetables sold in supermarkets from Ribeirão Preto, São Paulo, that came from that same region were also contaminated with intestinal parasites. All these observations offer relevant insights for public health to be rethought in light of the consumption of agricultural produce, expanding inspections in this sector with the goal of reducing diseases transmitted by produce that are naturally eaten raw. Water sanitation and the need for treatment prior to irrigation must be rethought to promote the quality of life of the population with a view to reducing infectious and parasitic diseases.

From a microbiological point of view, the data from our study indicate that the water evaluated is unsuitable for irrigating vegetables eaten raw, which requires the adoption of new basic sanitation measures to treat the sewage received by the STP built by the city government. Several studies indicate that food is a vehicle for pathogenic microorganisms, hence, the use of contaminated water for human activities results in a problematic that must be addressed by public health (Brazil, 2008).

The density of phytoplankton varied between 564 ind x mL⁻¹ (P1, March 2021) and 3376 ind x mL⁻¹ (P3, March 2021). Point P1, located near the source, presented a lower average density (1003 ind x mL⁻¹) than points P2 (1889 ind x mL⁻¹) and P3 (2289 ind x mL⁻¹), located downstream from the STP. The cyanobacteria *Aphanocapsa* sp. predominated in all three points, contributing for at least 80% of the total density. Species of the genus *Aphanocapsa* are on the list of picophytoplankton species that are deemed as potentially toxic (Jakubowska & Szeląg-Wasielewska, 2015). The higher contribution of *Cryptomonas* sp. (Cryptophyceae), an organism associated with meso-eutrophic environments, to the phytoplankton density at point P3 (Padisák, Crossetti & Naselli-Flores, 2009) is also noteworthy. These results show more favorable conditions for the development of phytoplankton at points P2 and P3, with a predominance of cyanobacteria, corroborating the evidence of the negative effect of the STP on the Corredor Stream's water quality.

Results obtained over the course of this research indicate the contamination of the body of water studied, which means that the water is unsuitable for both direct consumption and for irrigating vegetables produced by local farmers, sold on the streets, at farmers' markets, and eaten raw. Also, since the stream is contaminated, analyses of the surrounding groundwater are necessary to assess, for example, the damage to artesian wells located on the banks of the stream. If used for irrigation, the stream water may contaminate agricultural produce, which, in turn, may carry various pathogens to the consumer's table, posing a public health hazard for consumers.

CONCLUSION

The Corredor Stream in Mario Campos, Minas Gerais, has been regularly used by farmers in the surrounding area to irrigate vegetables eaten raw. However, there is no data in the literature to confirm the quality of the water in terms of the microbiological requirements set by Brazilian legislation. This study sought to evaluate the microbiological parameters required

by CONAMA Resolution no. 375/205 for the usage of water intended for irrigation, as mentioned.

Microbiological and physical-chemical analyses of the water from the Corredor Stream located in Mario Campos have thus demonstrated that this stream is contaminated by microorganisms from the coliform group, confirming *E. coli* both by the standard methods defined by the FUNASA Manual (Brazil, 2006) and by the presence of coliphages, and that some of its parameters are not in accordance with current regulations. The presence of a genus of cyanobacteria considered to be potentially toxic was also verified in higher densities at points downstream from the STP. Such water, therefore, should not be used for the irrigation of vegetables eaten raw. New proposals for basic sanitation complementary to the sewage treatment plant present in the region must be developed by the city government and applied to avoid contamination of food, groundwater and artesian wells used by the surrounding population.

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CONFLICTS OF INTEREST

The authors declare no conflict of interest.

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PEER REVIEW

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