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Benthic Macroinvertebrates as Bioindicators of Water Quality in the Vilcanota River, Cusco-Peru

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ABSTRACT

Macroinvertebrate metrics are excellent tools for assessing water quality due to the sensitivity of biotic and abiotic parameters of their environment. The work aimed to assess the water quality of the Vilcanota River using aquatic macroinvertebrates and biological indices: Andean Biotic Index (ABI), Biological Monitoring Working Party (BMWP) score, and Ephemeroptera, Plecoptera, and Trichoptera (EPT) index. Macroinvertebrates were sampled at four sampling points (P1, P2, P3, and P4) during dry and wet seasons using Surber traps along a 600 m linear transect. In total, 1631 specimens belonging to 04 classes, 11 orders, and 24 families were found. The class Insecta presented the highest values with 1078 specimens (66.1%), six orders (54.5%), and 19 families (79.2%). The evaluation of the water quality of the Vilcanota River showed that the points during the wet season have questionable water quality for the BMWP and ABI indices. In the dry season, most sampling points (except P1, classified as questionable) showed critical water quality in both the BMWP and ABI index. Similarly, the ETP index revealed regular water quality in the wet season, while in the dry season was bad water quality for most sampling (except P2) points.

1. INTRODUCTION

Rivers are important sources of freshwater that carry nutrients, microorganisms, and gases to many areas around the earth [1]. Their role is important in the water cycle, helps drain surface water, and provides habitats and food for many species of animals and plants [2]. Likewise, are vital for human life because offer many goods and services such as irrigation in agriculture, fishing, domestic uses, drinking water, transportation, leisure activities (swimming and boating), and electricity production [3, 4]. However, many river systems have been affected and degraded due to population increase and the development of several human activities such as mining, agriculture, industries, and domestic, whose effluents and residues may affect the water quality and aquatic ecosystems [5, 6].

Vilcanota River is one of the most important rivers in the Cusco Region because comprises Calca and Urubamba Provinces, which have a large population where the countryside and city coexist with a huge tourist current. This river still houses some fish, trout, otter, plankton, and several vegetables that serve as food. Vilcanota River is used by the surrounding community for several purposes such as agricultural activities, tourism, human consumption, animal husbandry, and amusement, and mainly because it supplies energy to the Machu Picchu power plant. However, there is a concern since the river receives all sewages generated by the population [7]. Likewise, there is a municipal dump installed on the left margin of the river, which is considered another source of pollution [8].

In this context, water quality may be evaluated using various biological, physical, or chemical indicators [9]. Benthic macroinvertebrates (BM) are small aquatic animals, primarily in their larval stages, that are commonly used as biological indicators to assess the condition of water bodies through their abundance and diversity [10]. The BM are reliable indicators because spend most or all their lives inside the aquatic system, respond to human disturbance, are easy to collect and identify in the laboratory, have limited mobility and capacity to integrate the effect of the stressors, and differ in their tolerance to pollution [10-12]. A high number in diversity and abundance of BM may indicate a healthy water

body. If the water body is healthy biologically, the physical and chemical components are also in good condition [10].

As rivers differ in geographical regions, distribution, biodiversity, and conditions, countries have developed and applied different biotic indices [13, 14]. For instance, in Peru water quality in several rivers was estimated using the ABI, EPT Index, BMWP Index, among others [15-17]. In Cusco Region studies using biotic indices are scarce, being that in the scientific literature only was reported a work that evaluated the water quality for the Amazon streams between Puerto Maldonado and Cusco [18]. Thus, the main objective of this study was to assess the water quality biologically from Vilcanota River using data about macrobenthic invertebrates as indicators through three biotic indices: i) ABI, ii) BMWP score, and iii) EPT index. The results found may be used for determining the management of the Vilcanota River Ecosystem.

2. METHODOLOGY

2.1 Study area and selection of sites

The Vilcanota Urubamba basin (73°47' and 73° 44' W, 14°39' and 10°09' S) is situated between the Cusco and Ucayali Department of Peru at an altitude of 2950 meters above sea

level. This basin presents three main drainage axes: Vilcanota, Mapacho, and Yanatile Rivers. The Vilcanota River is located in the Calca district and flows from north to east of the city of Cusco, Peru. The average annual temperature fluctuates between 11 and 16°C, the maximum between 22 and 29°C, and the minimum between 7 and -4°C during the winter [19]. Rainfall is regularly between December to March and drought between August to October.

2.2 Periods and sampling points

The sampling of benthic macroinvertebrates was carried out during the wet (November-December 2020 and January 2021) and dry (August to October 2021) seasons at four points: P1 (13°20'9.56" S; 71°57'23.90" W), P2 (13°19'51.50' S; 71°57'22.94" W), P3 (13°19"39.66" S; 71°57'17.39" W), and P4 (13°19'33.84" S; 71°57'36.21" W) (Figure 1). The points represent four different groups of land use (Figure 2): P1 is located 50m upstream of the discharge of the leachate from the dump in the Campanachoc sector; P2 to 200m downstream of the discharge of leachate from the municipal dump (ETSA); P3 is located to 100m upstream of the suspension bridge Calca, where people throw their garbage; and P4 located at 50m from the mouth of the Qochoq River. In conclusion, all points influence anthropic activities and discharges of solid and liquid pollutants.



Figure 1. Location of the sampling points along Vilcanota River, Cusco, Perú



Figure 2. Collection points of substrate samples along the Vilcanota River, Calca district, Cusco, Peru Source: Authors' photographs

2.3 Sampling and identification of benthonic macroinvertebrates

The benthic macroinvertebrates were collected following the methodology described in the book "Methods of Collection, identification, and Analysis of biological periphyton. communities: plankton. benthos (macroinvertebrates) and nekton (fish) in inland waters of Peru" [20]. Before sampling, the bottom was removed and covered an area of 6m². The samples were collected from downstream towards upstream to minimize the collection of drifted organisms. It was quantitatively carried out using a rectangular-shaped Surber net (50cm wide x 25cm high and 250µm pore size). Additionally, the organisms attached to stones, branches, leaves, and other objects that are in the place were collected. Collected macroinvertebrates were washed in the field and each sample separately was preserved in plastic bottles (500mL) using 70% ethanol [21, 22].

Simples were taken to the laboratory, where families were identified using the methodology described by Samanez et al. [20].

2.4 Data analysis

Descriptive analysis was carried out on the physicochemical parameters. Differences among sampling points were performed through the One-way ANOVA and post hoc Tukey Test. In addition, the Total Abundance (N) was calculated: total number of individuals collected at each sampling point; and Richness (S): total number of taxa recorded at each sampling point. Water quality was evaluated and compared using three biological indices: BMWP score, ABI, and Ephemeroptera, Plecoptera, and Trichoptera (EPT) index.

2.5 Biological Monitoring Working Party (BMWP) score

The BMWP is a simple, fast, and qualitative method (presence or absence) that allows to evaluation of the quality of the water using macroinvertebrates that only requires reaching the family level. The score ranges from 1 to 10 according to the tolerance of the different groups to organic pollution, with the most sensitive families receiving a score of 10, and the most tolerant only one. The sum of the scores of all families provides the BMWP total score (Eq. (1)) and its classification is given in Table 1.

$$BMWP = T1 + T2 + T3 + \dots Tn$$
 (1)

where, *T*=tolerance level of each macroinvertebrate found, and the number corresponds to the family.

Table 1. BMWP and ABI index classification of water quality according to Ríos-Touma et al. [13], respectively

Class	G Quality	BMWP Index	ABI Index	Meaning	Color
Ι	Good	>150 101- 120	>70	Very clean waters Unpolluted waters	
Π	Acceptable	61-100	45-70	Effects of contamination are evident	
III	Questionable	36-60	27-44	Waters moderately polluted	
IV	Critical	16-35	11-26	Waters heavily polluted	
V	Very critical	<15	<11	Waters severely polluted	

2.6 Andean biotic index (ABI)

The ABI method was calculated to assess the water quality and is based on the adaptations of the BMWP score for Andean areas with altitudes major than 2000 meters above sea level, being thus, widely used in Peruvian, Ecuadorian, and Colombian streams [13]. Its adaptation provides tolerance values for families of macroinvertebrates present in lotic environments whose tolerance values are from 1 (more tolerant) to 10 (more sensitive), and the total sum is the ABI score Eq. (2). The ABI classification is shown in Table 1.

$$ABI = T1 + T2 + T3... + Tn$$
 (2)

where, *T*=tolerance level of each macroinvertebrate found, and the number corresponds to the family.

2.7 Ephemeroptera, Plecoptera, and Trichoptera (EPT) index

The EPT index is a measure of the percentage of Ephemeroptera (mayflies), Plecoptera (stoneflies), and Trichoptera (caddisflies) which estimates water quality by the relative abundance of these three orders of stream insects that have low tolerance to water pollution [23]. It is one of the methods more useful and effective of the macroinvertebrate indices, especially in lotic ecosystems [24]. This method has been successfully used in other studies that evaluated the river water quality [25-27]. The EPT value represents the sum of the taxa richness of these three orders and is found by dividing the number of EPT by the total number of individuals Eq. (3) at the sampling point and is classified according to Table 2.

$$ETP = (number of \frac{EPT}{number} of individuals)^* 100$$
(3)

where, the number of EPT refers to the total of individuals of orders Ephemeroptera, plecopteran, and trichopteran, and the number of individuals represents the total registered of other types of orders or families in total.

Table 2. ETP index classification of water quality according
to Carrera and Fierro [28]

ETP Value (%)	Quality/Interpretation	Color
75-100%	Very Good	
50-74%	Good	
25-49%	Regular	
0-24%	Bad	

3. RESULTS

3.1 Physicochemical parameters

Table 3 shows the variations of physicochemical parameters of the Vilcanota River in the two periods of study: wet season (November-December 2020 and Janeiro 2021) and dry season (August to October 2021). The National Environmental Quality Standards for Water (ECA), Category III, Irrigation of vegetables and animal drinking through the Supreme Decree No 002-2008-MINAM was applied for comparison purposes (MINAM 2008) [29]. In this document, is presented the maximum permissible limits (MPL) for pH (6.5-8.5), temperature (<35°C), conductivity (<2000 μ S cm⁻¹), dissolved oxygen (\geq 4mg L⁻¹), and others.

The temperature ranged from 14.3 to 15.2° C in the wet season, with the lowest mean temperature (14.3°C) recorded at P3, while in the dry season varied from 16.1 to 16.6°C, with the lowest mean temperature (16.1°C) also measured at P3 (Table 3). Likewise, the highest temperature for each season was found at P4. On the other hand, no season or sampling points exceeded MPL. Between seasons significant differences (p>0.05) were observed.

The lowest conductivity value was registered at P3 during the wet season $(202\mu S \text{ cm}^{-1})$ and dry season $(1023\mu S \text{ cm}^{-1})$. Besides, it was observed a significant (p>0.05) increase in the mean conductivity during the dry season $(1100\pm 66\mu S \text{ cm}^{-1})$ compared to the wet season $(218\pm 14\mu S \text{ cm}^{-1})$. However, neither the wet season nor the dry season exceeded the MPL.

The pH levels in the dry season (7.5 ± 0.1) and wet season (7.6 ± 0.1) were within the recommended values (6.5-8.5) of the MPL. Likewise, no significant differences were observed between seasons and point sampling.

The dissolved oxygen (D.O.) ranged from 5.2 to 5.6mg L⁻¹ and 4.9 to 5.2mg L⁻¹ in the wet and dry seasons, respectively. The lowest DO (5.0mg L⁻¹) was recorded at P4 (dry season) and the highest (5.6mg L⁻¹) was recorded at P1 (wet season), and there were no significant differences among sampling points and seasons (p<0.05).

Table 3. Physicochemical parameters were measured on

 Vilcanota River samples collected during the wet and dry seasons

Parameter		Wet Season							
rarameter	P1	P2	P3	P4	Mean±S.D.				
T (°C)	14.7	15.2	14.3	14.9	14.8±0.3 a				
Conductivity (µS cm ⁻¹)	240	210	202	220	218±14 a				
pН	7.5	7.4	7.7	7.6	7.6±0.1 a				
D.O. (mg L ⁻¹)	5.6	5.2	5.5	5.3	5.4±0.2 a				
Parameter	Dry Season								
rarameter	P1	P2	P3	P4	Mean±S.D.				
T (°C)	16.2	16.5	16.1	16.6	16.4±0.2 b				
Conductivity (µS cm ⁻¹)	1200	1064	1023	1112	1100±66 b				
pН	7.5	7.5	7.4	7.5	7.5±0.1 a				
D.O. (mg L ⁻¹)	4.9	5.1	5.2	5.0	5.1±0.1 a				

T=temperature; D.O.=dissolved oxygen, S.D.=standard deviation The same letter on each line indicates no significant difference (p >0.05)

3.2 Diversity of benthonic macroinvertebrates

In total, 1631 individuals' belongings to the aquatic macroinvertebrate community were collected during the wet (882 individuals, 54%) and dry (749 individuals, 46%) seasons. They were organized into 04 classes, 11 orders, and 24 families (Table 4 and Figure 3). In the two seasons, the class Insecta represented best the point samplings, both in terms of order and families. This class was presented by 1078 specimens (66.1%), six orders (54.5%), and 19 families (79.2%). In general, the best-represented order in the sampling area was Diptera with 494 individuals (45.8%), and four families, Trichoptera with 333 specimens (30.9%), and five families, Odonato with 121 specimens (11.2%) and five families, Ephemeroptera with 52 specimens (4.8%) and one family, Coleoptera with 51 specimens (4.7%) and three families and last the Hemiptera with 27 specimens (2.5%) and one family. Besides, the class Clistellata (211 specimens, 12.9%) was represented by three orders: Haplotaxida (55 specimens, 26.1%), Hirudinea (107 specimens, 50.7%), and Oligochaeta (49 specimens, 23.2%), with one family each (Table 5).

In the wet season, the most abundant families were *Ceratopogonidae* (class Insecta, order Diptera) and *Hydropsychidae* (class Insecta, order Trichoptera) with a total of 276 and 95 individuals corresponding to 31.3% and 10.8%, respectively of the specimens collected. The dry season was represented by the family *Chironomidae* (Class Insecta, order Diptera) and the family *Physidae* (Class Gastropoda, order Sommatophora) with 196 and 192 individuals that corresponding to 26.2% and 25.6%, respectively of the specimens found. In function to the number of specimens, the wet season showed the following order: P4 (313 specimens) >P1 (216 specimens) >P3 (198 specimens) >P2 (155 specimens), similarly the dry season presented the following sequence: P4 (228 specimens) >P1 (191 specimens) >P3 (172 specimens) >P2 (158 specimens) (Table 5).

In function to the number of families (richness) during the wet season, the P1 showed the presence of 15 families (*Ceratopogonidae* the most abundant, n=62), followed by P2 and P3, both with 14 families (*Hydrobiosidae*, n=52 and *Ceratopogonidae*, n=67 the most abundant, respectively), and P4 with 11 families (*Ceratopogonidae* the most abundant, n=132). In the dry season was P1 with 13 families, followed by P2 and P4, both with 11 families, and P3 with 08 families.

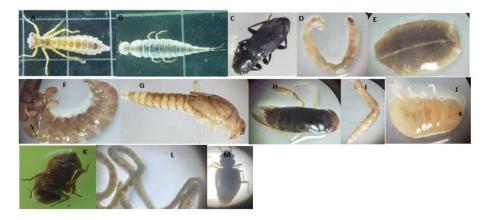


Figure 3. Some specimens found on the Vilcanota River: A: Familia Aeshnidae, B: Dysticidae, C: Stenelmis sp. (Elmidae), D: Familia Chironomidae, E: Turbellaria, F: Leptonema sp, G: Familia Baetidae, H: Corixidae, I: Aphrosylus sp T, J: Hyallela sp. (Hyalellidae), K: Curculionidae, L: Chordodidae, and M: Hydrophilidae

Table 4. Summary of the analysis of water quality using the BMWP/Col, and ABI indexes

		Wet S	eason	Dry Season					
	P1	P2	P3	P4	P1	P2	P3	P4	
BMWP	41	42	40	40	36	25	25	22	
Class	III	III	III	III	III	IV	IV	IV	
Quality	Questionable	Questionable	Questionable	Questionable	Questionable	Critical	Critical	Critical	
Meaning	Moderately	Moderately	Moderately	Moderately	Moderately	Heavily	Heavily	Heavily	
Wiedning	polluted	polluted	polluted	polluted	polluted	polluted	polluted	polluted	
Color									
ABI	34	36	38	38	28	24	23	18	
Class	III	III	III	III	III	IV	IV	IV	
Quality	Questionable	Questionable	Questionable	Questionable	Questionable	Critical	Critical	Critical	
Meaning	Moderately	Moderately	Moderately	Moderately	Moderately	Heavily	Heavily	Heavily	
wicannig	polluted	polluted	polluted	polluted	polluted	polluted	polluted	polluted	
Color									

 Table 5. Abundance and richness of aquatic macroinvertebrates from the sampling points in the Vilcanota River, during the wet and dry season

	0.1	E	Wet Season				Dry Season					T. 4.1			
Class	Order	Family	P1	P2	P3	P4	Total	%	P1	P2	P3	P4	Total	%	Total
		Mlathyria Marcella	10	02	-	08	20	2.3	-	-	-	-	-	-	
		Acanthagrion	07	02	02	-	11	1.2	-	-	-	-	-	-	
	Odonata	Aeshnidae	03	04	-	01	08	0.9	12	-	-	01	13	1.7	121
		Gomphidae	-	-	-	-	-	-	05	02	07	05	19	2.5	
		Libellulidae	28	-	13	09	50	5.7	-	-	-	-	-	-	
		Hydropsychidae	09	24	16	46	95	10.8	11	-	-	04	15	2.0	
		Helicopsychidae	11	02	02	07	22	2.5	-	34	11	-	45	6.0	
	Trichoptera	Philopotamidae	23	02	-	45	70	7.9	-	-	-	-	-	-	333
	-	Polycentropodidae	-	-	24	-	24	2.7	-	-	-	-	-	-	
Insecta		Hydrobiosidae	-	52	10	-	62	7.0	-	-	-	-	-	-	
		Hydrophilidae	-	-	-	-	-	-	05	02	05	01	13	1.7	
	Coleoptera	Elmidae	-	-	-	-	-	-	-	01	04	-	05	0.7	51
	•	Dytiscidae	-	-	-	-	-	-	33	-	-	-	33	4.4	
	Ephemeroptera	Leptohyphidae	14	08	16	-	38	4.3	04	05	06	-	15	2.0	52
		Chironomidae	-	-	-	-	-	-	48	40	75	33	196	26.2	
	D' (Ceratopogonidae	62	15	67	132	276	31.3	-	-	-	-	-	-	40.4
	Diptera	Empididae	-	-	-	-	-	-	05	03	-	-	08	1.1	494
		Aphrosylus	07	05	01	-	13	1.5	01	-	-	-	01	0.1	
	Hemiptera	Pleidae	06	06	03	02	17	1.9	03	05	-	02	10	1.3	27
	Haplotaxida	Tubificidae	02	06	04	-	12	1.4	-	-	-	43	43	5.7	
Clitellata	Hirudinea	Glossiphoniidae	-	-	-	-	-	-	10	12	32	53	107	14.3	211
	Oligochaeta	Oligochaeta	08	14	12	06	40	4.5	-	-	-	09	09	1.2	
Malacostraca	Amphipoda	Hyalellidae	07	-	21	34	62	7.0	14	01	-	10	25	3.3	87
Gastropod	Basommatophora	Physidae	19	14	07	23	63	7.1	40	53	32	67	192	25.6	255
-	•	Abundance (N)	216	155	198	313	882	100	191	158	172	228	749	100	1631
		Richness (S)	15	14	14	11	17		13	11	08	11	16		

Table 6. Summary of the analysis of water quality using the ETP index

Sampling	Season	EPT I	Color	
Point	Season	Value (%)	Quality	Color
D1	Wet	26	Regular	
P1	Dry	8	Bad	
P2	Wet	56	Good	
P2	Dry	25	Regular	
P3	Wet	34	Regular	
P3	Dry	10	Bad	
P4	Wet	31	Regular	
P4	Dry	2	Bad	

3.3 Evaluation of water quality

Table 4 presents a summary of the water quality results after applying the BMWP/Col, and ABI indexes in each sampling point and season.

For the BMWP index, all sampling points in the wet season showed moderate values (ranging from 40 to 42), indicating that the water quality in each sampling point is questionable (moderately polluted), while most sampling points (except P1) in the dry season presented lower values (ranging from 22 to 25), suggesting that the water quality is critical (heavily polluted) (Table 4).

The ABI index reported similar findings and water quality the BMWP index, with questionable quality during the wet season and most sampling points during the dry season presented a water quality as critical. Of all sampling points, P4 was who presented the lower values, indicating that this point is the more susceptible to pollution. Besides, the decrease of families is evident, especially in the orders *Ephemeroptera* and *Trichoptera*, as well as the presence of macroinvertebrates more tolerant to pollution, such as those of the order Diptera (especially in the sampling point P4).

Table 6 shows a summary of the water quality using the ETP index at each sampling point and season. As observed in Table 6, in the wet season all sampling points presented water quality classified as regular (except P2 - good), while in the dry season, most sampling points indicated water quality as bad (except P2-regular). Here is possible note that P1 (8%) and P4 (2%) were those that presented lower values (%), suggesting that they are the points that probably suffer the greatest impact of pollution.

4. DISCUSSION

4.1 Physicochemical parameters

The physicochemical parameters showed relatively similar values (except the temperature and conductivity) for most sampling points and seasons.

In the temperature, both seasons did not exceed the MPL, indicating that the river complies with these specifications. A slight difference in mean temperature among sampling points and seasons (14.8±0.3°C wet season and 16.4±0.3°C dry season) was observed. Similar findings were reported by Malakane et al. [30] measured the temperature at seven sampling points from the Blyde River (South Africa). found a variation of 22.3°C to 24.90°C. Likewise, Izatti and Retnaningdyah [31] in the Genjong River (Indonesia) found a temperature ranging from 16.7°C to 23.7°C at four different stations. Bonacina et al. [32] indicated that temperature is an abiotic factor that affects the structure and functioning of aquatic ecosystems and biological communities. Moreover, temperature influences the solubility of gases, pollutants, pH, electrical conductivity, density, and toxicity of chemicals. Higher temperatures promote Thus, an increase or decrease in temperature based on the season, climatic conditions, or changes in river flow could alter the number of biological communities. Likewise, changes in river flow of water [33]. In this work, the lowest temperature found may be related to a high altitude (colder air temperature) of the study area and to a decrease in the flow of water during the dry season.

The conductivity presented lower values in the wet season $(218\pm14\mu \text{S cm}^{-1})$ compared to the dry season $(1100\pm66\mu \text{S cm}^{-1})$. An increase or decrease in conductivity indicates pollution [34]. When fresh water is lost by evaporation the water level decreases, then the ions present become concentrated, and as a consequence, the conductivity increases [35]. This description may explain because a high conductivity level was found at all sampling points during the season. Besides, rivers less disturbed frequently present low conductivity, silt load, and turbidity [30]. The Vilcanota River showed moderate temperature, but higher levels of conductivity (202 to 1200 μ S cm⁻¹) compared to studies reported in the Sardinas River (Choco Andino, Ecuador) (4.4 to 22.0\mu S cm⁻¹) [36], and the Genjong River (Indonesia) (83.4 to 109\mu S cm⁻¹) [30].

The pH level of the water in wetlands, lakes, and rivers is an environmental factor key that limits species distribution and life in aquatic habitats because affects most biological and chemical processes [37]. The EPA recommends a range of pH optima between 6.5 to 8 for the development of aquatic organisms. Thus, as our pH measured in both seasons ranged from 7.4 to 7.6, this may be categorized as normal to slightly basic, being suitable for the life of aquatic organisms [38].

All aquatic life depends on the availability of dissolved oxygen (free oxygen present in water) [39]. In the wet (5.4 ± 0.2) and dry (5.1 ± 0.1) seasons the values of O.D. were higher as recommended by the MPL (\geq 4mg L⁻¹), which indicates that this water can contain life. Fonseca and Salvador [40] reported a reduction of O.D. concentration in the water when the temperature was increased during the rainy season. In our study, the dry season presented higher temperatures (16.1°C to 16.6°C) and a minor concentration of D.O (4.9 to 5.2mg L⁻¹) compared to the rainy season. Therefore, it was expected that in locations where the water temperature is higher, lower concentrations of D.O. would be found.

4.2 Macroinvertebrates

The Vilcanota River is one of the main rivers of the Cusco Region, which reaches the confluence of the Yanatile River, its section is impacted by various problems. Thus, it is necessary to carry out several actions of surveillance, monitoring, and control of the quality of water resources, to prevent, mitigate, and control impacts.

The number of macroinvertebrates found in the study area differed between seasons, being that the wet season (882 specimens) presented a major number of specimens compared to the dry season (749 specimens). Similar found were reported by Machado et al. [36] and Pascual et al. [16] reported significant changes in the composition of the macroinvertebrate community, with a greater number of specimens in the dry season than in the wet season, which coincides with our finding in the present study.

Based on the monitoring of macroinvertebrates, the family Insecta was the more representative in all sampling points and seasons. It was found that during the rainy season, *Ceratopogonidae* was dominant on P1 and P3, while *hydropsychidae* was dominant on P2 and P4 sampling points. Many species of *Ceratopogonidae* live in aquatic habitats such as ponds, freshwater marshes, swamps, lakes, and streams. During the rainy season are formed streams that favor the presence of this species, since during the dry period the presence of this species was not observed. Similarly, the *hydropsychidae* live in a wide range of lotic habitats from small streams to large rivers, lakes, and permanent or temporary ponds.

In the dry season, *Chironomidae* was dominant in all sampling points. The family *Chironomidae* is tolerant to water with low dissolved oxygen and a high concentration of conductivity [41, 42]. Our results reported lower levels of DO and higher values of conductivity, which would explain the presence of this family during the dry season. Likewise, several authors have associated its presence with a reduction in water quality and degradation of aquatic ecosystems [43, 44].

In rivers, lakes, creeks, and other freshwater habitats, insects are the most abundant and often exhibit a high diversity of macroinvertebrates in the benthic community [45]. Aquatic insects are derived from a variety of terrestrial ancestors and

play a significant role in freshwater ecosystems because serve as food for other species and are used as water-quality condition indicators of both lotic and lentic systems [46]. Similar findings were reported by other authors where the major class was Insecta [30, 36, 47]. Thus, our findings are understudies previously published.

In function to the order, the macroinvertebrates presented the following sequence: Diptera > Trichoptera > Basommatophora (class Gastropod) > Odonata, > Hirudines (class Clitellata) > Amphipoda (class Malacostraca) > Haplotaxida (class Clitellata) > Ephemeroptera > Coleoptera > Oligochaeta (class Clitellata) > Hemiptera.

The Diptera order was represented by the family Ceratopogonidae (276 individual specimens-biting midges) during the wet season and the family Chironomidae (196 individual specimens) represented the dry season. The biting midges are considered an important vector of human and veterinary pathogens and their population is increasing due to that rainfall leads to more hatching of mosquito eggs [48], the reason why in the rainy season only this family was found.

The order Trichoptera (Caddisflies) are aquatic insects, and most species are moderately tolerant to water pollution. A greater number of families of Trichoptera were found in the wet season compared to the dry season. These macroinvertebrates often tend to adapt to changes that allow survive in water with low dissolved oxygen or turbid waters indicating that only these organisms can survive in this stream.

The order Basommatophora is abundant, widespread, and known to be very tolerant of pollution, especially in water polluted with organic pollution [49]. In this case, this family was widely distributed in both seasons, which indicates that the water of the Vilcanota River is polluted. Rivers, lakes, or ponds with good water quality usually showed a high number of Odonata species [50]. Here, the wet season showed 89 specimens, while the dry season 32 specimens. Thus, this finding suggested that the water quality from the Vilcanota River is not good. The order Ephemeroptera (mayflies) is considered a good indicator of water quality, although some specimens can tolerate a certain degree of pollution [51]. Here, only 52 specimens (38 and 15 specimens in wet and dry seasons, respectively) were found, which may indicate bad or regular water quality. The order Plecoptera lives mainly in temperate and cold areas and well-oxygenated water running, being used as indicators of high water quality due to that these are extremely sensitive to water pollution [52]. In this study no family of the order Plecoptera was found, indicating pollution in the Vilcanota River water.

It is important to note that in the habitats, some taxa may not be present because of a migration pattern, seasonal variation, or unstable habitats.

4.3 Water quality

The water quality evaluated in each sampling point with the indexes BMWP and ABI showed that the waters of the Vilcanota River were classified as questionable (moderately polluted) for all sampling points during the wet season and P1 during the dry season, while critical (heavily polluted) for P2, P3, and P4 of the dry season. Like all sampling points were located around urban sites, the water quality of Vilcanota River assessed through macroinvertebrates is responded to and discriminated by the sites, where multiple stressors occur. In our case, the sites studied have influence such as increasing urbanization, agriculture, cattle farming, and presence of

municipal dumps, and the discharge of both solid and liquid pollutants on the Vilcanota River stream. Similar findings were reported by Castillejo et al. [53] in Ecuadorian Andean rivers.

Jerbes-Cobo et al. [54] indicated that ABI is primarily associated with variables such as DO, nitrite, and total solids during the rainy season. In this study, DO presents major values during the rainy season, indicating similar results and behavior. Likewise, high ABI values between 18 and 38 points indicate that these are influenced by the seasonality of precipitation and the flow of the body of water.

Besides, there is observed a decrease in water quality in the dry season compared to the wet season.

Similar findings were reported by Jacobsen and Encalada [55] who assessed eight bodies of water around the city of Ecuador and found high pollution during the dry season. Besides, the Alambi River basin (Ecuador) showed a decrease in the water quality index during the dry season [56]. Likewise, Pascual et al. [16] evaluated the water quality from the Rimac River and found a greater number of points polluted in the dry season than in the wet season.

These results of the ABI and BMWP index coincide with Coayla-Peñaloza [57] mention that its results were of regular generation in water quality and depends on different types of ecosystems of the place and the altitude, for which, the sandy bottoms are of great diversity, but the stony bottoms are of the great variety of supplies, in addition to the climatic conditions. They found a greater abundance of anthropic generations despite the wet season, their results showed greater sensitivity where they showed the taxonomic groups are some resistant to pollution.

The water quality of rivers is not static. Thus, river water quality changes constantly in response to environmental and human pressures, which display a dynamic variation over time [54].

As far as uniformity is concerned, all the points recorded in the Vilcanota River are given high values and polluted. Concerning the study carried out by Nuñez and Fragoso-Castilla [58] in the Ciénaga Mata de Palma River where the stations registered high values, which indicates that the population of aquatic macroinvertebrates is distributed homogeneously on the surface and bottom of the swamp. Espinosa et al. [59] stated that the distribution of aquatic macroinvertebrates in lentic ecosystems is mainly affected by the presence of floating vegetation, which constitutes a substrate and refuge for a great diversity of benthic communities.

Bad quality of the waters of the Vilcanota River then may be associated with the indiscriminate dumping of garbage and discharges of domestic and industrial wastewater as reported by the National Water Authority (ANA) and the Regional Government of Cusco [60]. Likewise, the ANA and local authorities of Cusco carried out actions to monitor and evaluate the quality of water and thus identified the main factors that affect the quality of the main river and its tributaries, as well as the main pressures that influence the watercourses that make it up. For this, a diagnosis study "Environmental Quality Index of Surface Water Resources (ICARHS)" was developed between 2012 to 2021, considering 143-point samplings through the Urubamba hydrographic units, where were measured physicochemical parameters (D.O., DQO, DBO₅,), trace elements (Pb, Mn, Zn, Fe, Al), and microbiological parameters (thermotolerant coliforms), revealing that 43 (30%) sampling points were classified as lousy, 54 (38%) as bad, 38 (27%) were qualified as regular and only 8 (6%) as good. According to our findings, the Vilcanota River had an important influence and pressure on anthropogenic activities along its course. Baytaşoğlu and Gözler [61] after the application of BMWP and ETP index concluded that urbanization, tourism, agricultural activities, and destruction of the river are the main responsible for bad water quality from Coruh River Basin (Turkey). In this study, we can say that all the reasons described above may have an effective role in the streams of the Vilcanota River.

The EPT index defines signs of bad quality overall in the dry season and regularly during the wet season, which are practically related to the small sample of specimens of EPT of families collected, and also to that was no found the order Plecoptera, which may explain the generation of pollution in the place by the discharged waters and generations of waste by the same community.

5. CONCLUSIONS

This study is a contribution to the knowledge of benthic macroinvertebrates in the Vilcanota River where was found a great abundance and richness of specimens allowed to be established. The BMWP index and ABI indexes revealed that the water of Vilcanota River was classified as questionable (moderately polluted) in all (P1 to P4) points during the wet season. In the dry season, both indexes classified the P1 as questionable (moderately polluted) and the P2, P3, and P4 as Critical (heavily polluted). Likewise, the ETP index classified the water of Vilcanota River as Regular for most points samplings (except P2 classified as good) during the wet season. In contrast, during the dry season, most points (except P2 as regular) were classified as Bad. In general terms, water quality based on three indices indicates that the Vilcanota River presents water moderately polluted during the wet season and heavily polluted in the dry season.

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