

ASSESSMENT OF TROPHIC STATUS AND WATER QUALITY IN THE LITTORAL AND PELAGIC ZONES OF LAKE OHRID BASED ON PHYSICO-CHEMICAL PARAMETERS

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Abstract: Lake ecosystems are highly dependent on the characteristics and processes occurring within their surrounding catchment areas, which directly influence the physical, chemical, and biological properties of the receiving water bodies. Anthropogenic pressures and natural processes within the catchment can substantially affect nutrient inputs and consequently influence water quality and trophic conditions. Increased nutrient availability may induce changes in the trophic status of the littoral zone, which may subsequently affect the ecological conditions of the pelagic zone. The trophic status of an aquatic ecosystem reflects its level of biological productivity and provides an important indication of its ecological condition. Lake Ohrid is among the most important natural freshwater ecosystems in North Macedonia and is globally recognized as one of the oldest lakes, supporting exceptionally diverse and endemic biota. Despite its exceptional ecological value and predominantly oligotrophic character, Lake Ohrid is not immune to the ongoing pressures associated with anthropogenic activities and the progressive process of eutrophication. Anthropogenic influence is particularly evident in certain parts of the shoreline, where localized areas of increased pressure or so-called “hot spots” may occur and potentially affect water quality and trophic conditions.

The aim of this study was to assess water quality and trophic status in the littoral zone of Lake Ohrid, with particular emphasis on areas affected by anthropogenic activities, and to provide a comparative assessment with a selected site in the pelagic zone. Seasonal sampling was conducted during 2025 and 2026 at six littoral sites distributed along the Lake Ohrid shoreline and at one pelagic sampling site, where a vertical profile comprising eight depths was investigated. Water samples were collected, stored and handled according to standard procedures prior to laboratory analyses. The investigated physico-chemical parameters included concentration of dissolved oxygen, biochemical oxygen demand (BOD₅), oxidizable organic matter expressed as KMnO₄ consumption, concentration of total phosphorus (TP), and total nitrogen (TN). Standard limnological methods were applied for the determination of the selected parameters. The trophic status of the investigated sites was evaluated using Carlson's Trophic State Index (TSI), the OECD classification, and the national Directive for Classification of Surface Waters. The results revealed pronounced spatial differences in the degree of anthropogenic influence among the investigated littoral sites. The concentrations of biodegradable organic matter ranged from 4.578 mg L⁻¹ KMnO₄ at the Desaret site to 12.597 mg L⁻¹ KMnO₄ at the Grashnica littoral and 12,929597 mg L⁻¹ KMnO₄ at Euro Hotel. The highest concentrations of total phosphorus were recorded at the Grashnica littoral (18.29 µg L⁻¹ TP) and the Sateska River littoral (20.15 µg L⁻¹ TP), indicating a pronounced influence of inflowing waters. Regarding the Trophic State Index based on total phosphorus, the lowest value was recorded in the pelagic zone (TSI-TP = 28), whereas the highest values were recorded at the Grashnica (TSI-TP = 46) and Sateska River littoral (TSI-TP = 47) sites. The application of Carlson's method, based on total phosphorus (TP) concentration and Secchi disk transparency (SD), provided numerical values of the trophic state index and enabled the classification of the investigated sites according to their trophic conditions. The obtained values indicate that the littoral zone of Lake Ohrid ranged from oligotrophic to oligo-mesotrophic conditions, whereas the pelagic zone corresponded to an oligotrophic state (average TSI-SD=21). The observed spatial variability indicates that localized anthropogenic pressures can result in measurable differences in physical and chemical conditions and trophic status within different parts of the lake. Overall, the findings demonstrate that river mouths, agricultural areas, and intensively used tourist zones represent important localized sources of anthropogenic pressure on the littoral zone of Lake Ohrid. The results confirm the importance of continued and systematic monitoring of both littoral and pelagic zones, integrating physical, chemical and biological parameters, together with targeted management measures aimed at reducing anthropogenic pressures, preventing further deterioration of water quality, and safeguarding the ecological integrity and long-term sustainability of this unique lake ecosystem.

Keywords: anthropogenic pressure, water quality, eutrophication, trophic status, littoral zone

1. INTRODUCTION

Lake ecosystems are dynamic systems whose physical, chemical, and biological conditions are closely linked to processes occurring within their catchments. Nutrient inputs from natural and anthropogenic sources, hydrology, land use, urbanization, agriculture, and wastewater discharges can substantially affect water quality and ecosystem productivity. Assessment of trophic status is therefore an important component of lake monitoring

and eutrophication management (Veljanoska-Sarafiloska et al., 2019; García-Ávila et al., 2025). Nitrogen and phosphorus are particularly important because increased loading can stimulate primary production, alter aquatic vegetation, and affect water quality and biodiversity (Matzinger et al., 2004, 2006; Vermaat et al., 2020; Schneider et al., 2020; Riza et al., 2023).

Trophic status integrates the productivity of a water body and is commonly evaluated using total phosphorus, chlorophyll *a*, and water transparency. Carlson (1977) developed the Trophic State Index (TSI) to allow quantitative comparisons among lakes and among areas within the same lake. The OECD classification similarly distinguishes ultra-oligotrophic, oligotrophic, mesotrophic, eutrophic, and hypertrophic conditions on the basis of nutrient and productivity-related variables (OECD, 1982). Oligotrophic lakes are generally characterized by low nutrient concentrations, high transparency, and good oxygenation, but even relatively small increases in nutrient loading may have important long-term consequences in sensitive systems.

Lake Ohrid, situated on the border between North Macedonia and Albania, is one of Europe's ancient lakes and is characterized by exceptional biodiversity and a high degree of endemism (Albrecht and Wilke, 2008). It is a deep, large-volume lake with a naturally oligotrophic state. Because of its outstanding natural and cultural value, the Ohrid region has been included on the UNESCO World Heritage List since 1979 for its natural values and since 1980 as a mixed natural and cultural property; in 2019, the protected property was extended to include the Albanian part of the lake. UNESCO recognizes the lake as an important refuge for more than 200 endemic plant and animal species and emphasizes the importance of preserving its oligotrophic conditions.

Despite these characteristics, Lake Ohrid is exposed to increasing anthropogenic pressures associated with shoreline urbanization, municipal wastewater, agriculture, tourism, and nutrient inputs through tributaries and groundwater. Long-term studies have identified phosphorus enrichment and changes in deep-water oxygen conditions as major ecological concerns. Matzinger et al. (2007) estimated an approximately 3.5-fold increase in phosphorus concentration in Lake Ohrid over the previous century. Although the lake remained oligotrophic, increased phosphorus loading was identified as an important long-term threat because enhanced organic matter production and mineralization may increase oxygen consumption in the deep waters.

The littoral zone is particularly important because it represents the interface between catchment-derived inputs and processes occurring within the lake. Vermaat et al. (2020) estimated that approximately 73% of external phosphorus loading originates from domestic point sources, while littoral vegetation retains about 31% of the phosphorus entering this zone. This confirms the important role of the littoral zone as a biogeochemical buffer between the catchment and the open lake. At the same time, littoral biological indicators may reveal nutrient enrichment even when pelagic nutrient concentrations remain low (Schneider et al., 2014, 2020).

The aim of this study was to assess the water quality and trophic status of Lake Ohrid using selected physicochemical parameters, with particular emphasis on spatial differences between littoral sites exposed to varying anthropogenic pressures and the pelagic zone. An additional objective was to identify localized areas of increased organic and nutrient loading and to evaluate their relevance to the preservation of the oligotrophic character of Lake Ohrid.

2. MATERIALS AND METHODS

The study was conducted in Lake Ohrid during seasonal sampling campaigns in 2025 and 2026. In the graphs for total nitrogen and total phosphorus, in addition to the seasonal data, the mean values from the four seasonal campaigns (which include two replicates), are also presented. Six littoral sampling sites were investigated: Euro Hotel, Sateska River littoral, Grashnica, Mazija, Konjsko, and Desaret. The selected sites represent shoreline sectors exposed to different potential pressures, including river inflows, populated and tourist areas, and sites with lower direct anthropogenic influence. A representative pelagic station was also investigated at eight depths: 0.5, 10, 20, 30, 40, 50, 100, and 150 m.

Littoral water samples were collected using a Ruttner water sampler (Hydro-Bios, Kiel, Germany), while samples from the pelagic vertical profile were collected using 5-L Niskin bottles. Sampling and analytical procedures followed standard methods (APHA/AWWA/WEF, 2012).

Water transparency was measured with a Secchi disk. Dissolved oxygen (DO) was determined iodometrically using the Winkler principle (ISO 5813:1983; APHA/AWWA/WEF, 2012). Biochemical oxygen demand over five days (BOD₅) was determined from the difference between the initial DO concentration and the concentration measured after five days of incubation at 20 °C in the dark. Organic matter was estimated by permanganate consumption (KMnO₄ consumption), using acidic digestion followed by titration. This parameter was used as an indicator of organic loading rather than as a direct measure of biodegradable organic matter. Total phosphorus (TP) was determined after persulfate digestion at 121 °C for 1 h, followed by spectrophotometric determination of orthophosphate using the ammonium molybdate-antimony method and ascorbic acid reduction (ISO 6878:2004). Total nitrogen (TN) was determined from the measured inorganic and Kjeldahl nitrogen fractions (TN = TKN + NO₂-N + NO₃-N), according to Standard Methods (APHA/AWWA/WEF, 2012).

Water-quality classes were interpreted according to the Regulation on Water Classification (Official Gazette of the Republic of Macedonia No. 18/99). Trophic state was additionally evaluated using Carlson's Trophic State Index based on total phosphorus and Secchi disk transparency (Carlson, 1977), with trophic interpretation considered together with the OECD framework (OECD, 1982). For total phosphorus concentration expressed in $\mu\text{g L}^{-1}$, the index was calculated according to:

$$\text{TSI}(\text{TP})=14.42\ln(\text{TP})+4.15$$

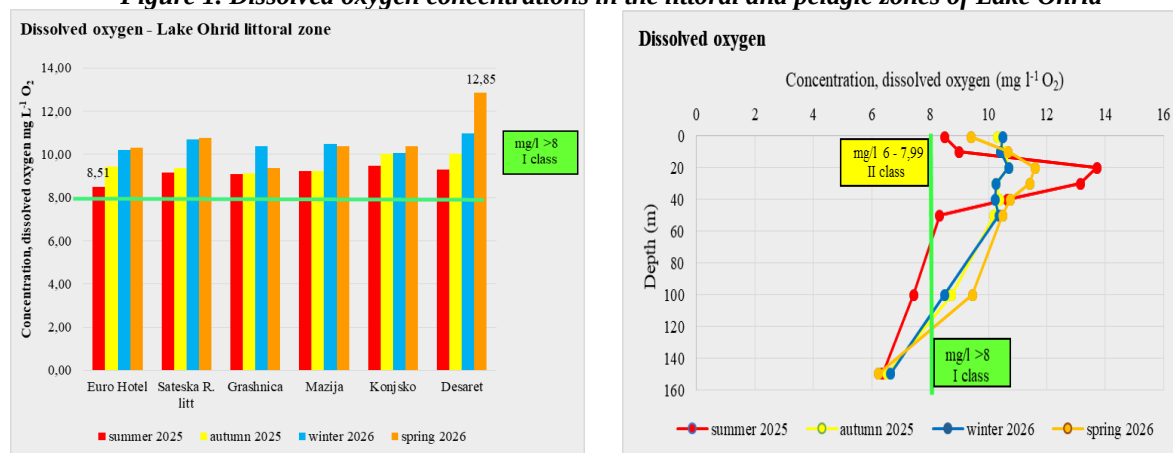
For Secchi disk transparency expressed in metres:

$$\text{TSI}(\text{SD})=60-14.41\ln(\text{SD})$$

3. RESULTS AND DISCUSSION

3.1. Dissolved oxygen (DO) is an important indicator of water quality and ecological condition because it reflects the balance between atmospheric exchange, photosynthesis, respiration, and decomposition of organic matter in aquatic ecosystems (Wetzel and Likens, 2000). The dissolved oxygen concentrations measured in the littoral and pelagic zones of Lake Ohrid are presented in Figure 1.

Figure 1. Dissolved oxygen concentrations in the littoral and pelagic zones of Lake Ohrid



Source: Author's research

In the littoral zone, all investigated sites were well oxygenated, with DO concentrations above $8 \text{ mg L}^{-1} \text{ O}_2$. According to the Regulation on Water Classification (Official Gazette of the Republic of Macedonia No. 18/99), these values correspond to Class I water quality. Concentrations ranged from $8.51 \text{ mg L}^{-1} \text{ O}_2$ at Euro Hotel to $12.85 \text{ mg L}^{-1} \text{ O}_2$ at Desaret. The pelagic water column was also well oxygenated, particularly between 0.5 and 100 m. DO gradually decreased toward deeper layers, reaching a minimum of $6.22 \text{ mg L}^{-1} \text{ O}_2$ at 150 m, corresponding to Class II water quality. Seasonal variability was evident in both littoral and surface pelagic waters, with generally higher DO concentrations during winter and spring. This pattern is consistent with the greater solubility of oxygen at lower water temperatures. The decrease observed in deeper layers may reflect reduced photosynthetic production, oxygen consumption during respiration and decomposition of organic matter, and restricted oxygen replenishment during periods of thermal stratification.

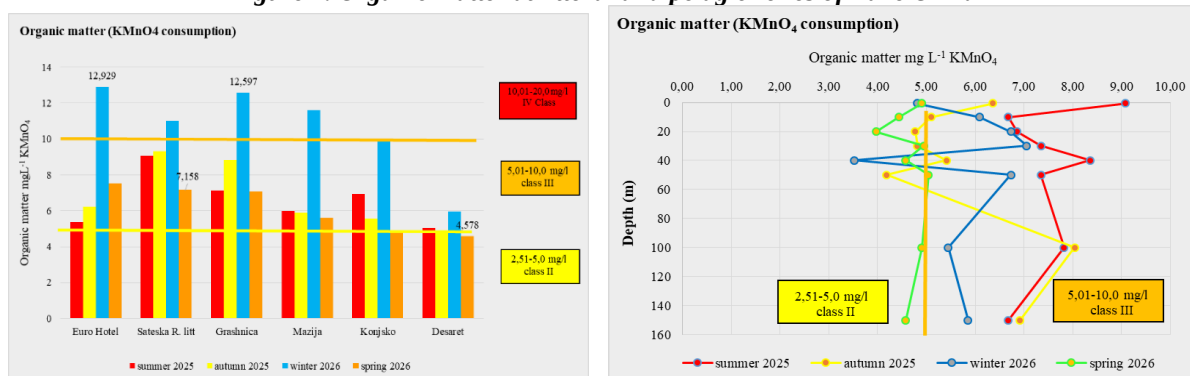
Similar processes have been reported in other lake ecosystems. Jane et al. (2021) documented widespread declines in dissolved oxygen in temperate lakes, relating surface-water oxygen losses mainly to increasing water temperature and reduced oxygen solubility, whereas deep-water declines were associated with stronger thermal stratification and changes in water clarity. For Lake Ohrid specifically, Matzinger et al. (2006, 2007) emphasized the sensitivity of deep-water oxygen conditions to nutrient enrichment and reduced vertical mixing. Their studies showed that increased phosphorus loading may enhance organic matter production and subsequent mineralization, thereby increasing oxygen consumption in deeper waters.

Overall, the present results indicate favorable oxygen conditions in both the littoral and pelagic zones of Lake Ohrid, with no evidence of severe oxygen depletion during the investigated period. Nevertheless, the decrease in DO toward deeper waters highlights the importance of continued monitoring, particularly in relation to nutrient loading, organic matter decomposition, and changes in lake stratification.

3.2. Organic matter and biochemical oxygen demand (BOD₅) provide complementary information on organic loading and oxygen-consuming processes in aquatic ecosystems. BOD₅ reflects the oxygen consumed during microbial degradation of biodegradable organic matter, whereas KMnO₄ consumption provides an estimate of oxidizable organic substances present in the water. Increased values of these parameters may therefore indicate enhanced organic inputs and microbial activity.

The concentrations of organic matter in the littoral and pelagic zones of Lake Ohrid are presented in Figure 2.

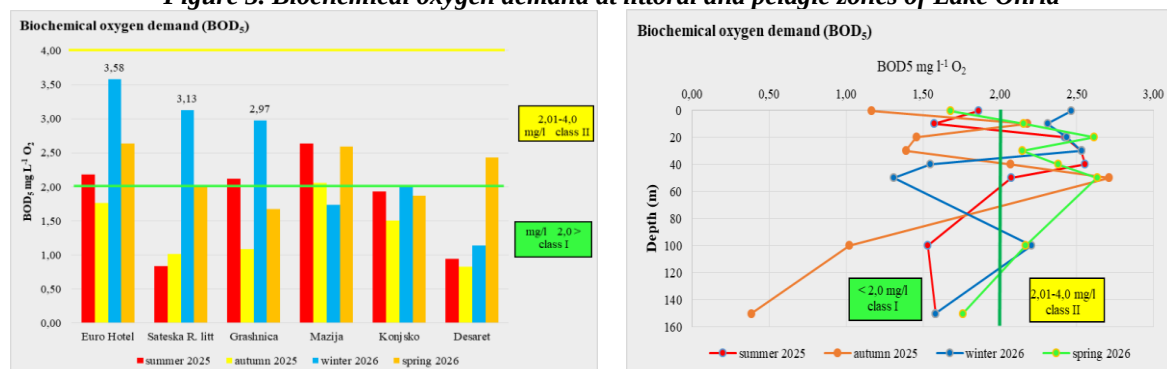
Figure 2. Organic matter at littoral and pelagic zones of Lake Ohrid



Source: Author's research

Clear spatial and seasonal variability was observed, with generally higher and more heterogeneous values in the littoral zone. The highest concentrations were recorded during winter 2026 at Euro Hotel (12.929 mg L⁻¹ KMnO₄) and Grashnica (12.597 mg L⁻¹ KMnO₄). In the pelagic zone, concentrations were more moderate, with a maximum of 9.08 mg L⁻¹ KMnO₄ in the surface layer (0.5 m) during summer 2025, and no consistent depth-dependent pattern was observed. The results for BOD₅ are presented in Figure 3.

Figure 3. Biochemical oxygen demand at littoral and pelagic zones of Lake Ohrid



Source: Author's research

Littoral values ranged from approximately 0.8 to 3.58 mg L⁻¹ O₂, with the highest concentrations recorded during winter 2026 at Euro Hotel (3.58 mg L⁻¹ O₂), Sateska River littoral (3.13 mg L⁻¹ O₂), and Grashnica (2.97 mg L⁻¹ O₂). In the pelagic zone, BOD₅ remained relatively low to moderate throughout the vertical profile (approximately 0.4–2.7 mg L⁻¹ O₂). The generally similar spatial pattern of organic matter and BOD₅ in the littoral zone suggests that sites with increased organic loading also tend to exhibit greater microbial oxygen demand.

Previous investigations of Lake Ohrid support this spatial pattern. Veljanoska-Sarafiloska et al. (2019) reported a positive relationship between average BOD₅ and organic matter concentrations during 2013–2014, with the highest average values of both parameters recorded at the Grashnica littoral site. Their comparative study classified Lake Ohrid as oligotrophic, whereas Lake Prespa showed mesotrophic conditions with a tendency toward eutrophication. In another study, Lokoska et al. (2019) recorded a KMnO₄-based organic matter value of 16 mg L⁻¹ at Grashnica in summer 2014 and substantially higher concentrations in the inflowing Velgoshka River (45 mg L⁻¹). These results were associated with municipal and other anthropogenic inputs transported by tributaries, indicating that Grashnica has historically represented an area of increased organic pressure. Similar relationships between organic matter, BOD₅, and trophic state have been observed in other lake systems. Yang et al. (2024), studying 68 lakes ranging from oligotrophic to eutrophic conditions, demonstrated significant relationships between dissolved organic matter characteristics and both CODMn and BOD₅, confirming their complementary value in assessing organic loading. Likewise, Liu et al. (2022) found higher organic matter levels in eutrophic than in oligotrophic lakes. At the European scale, Vigiak et al. (2019) emphasized BOD as an indicator of organic pollution and identified domestic wastewater, livestock waste, industrial emissions, and sewer overflows as important anthropogenic sources of organic loading to freshwater ecosystems.

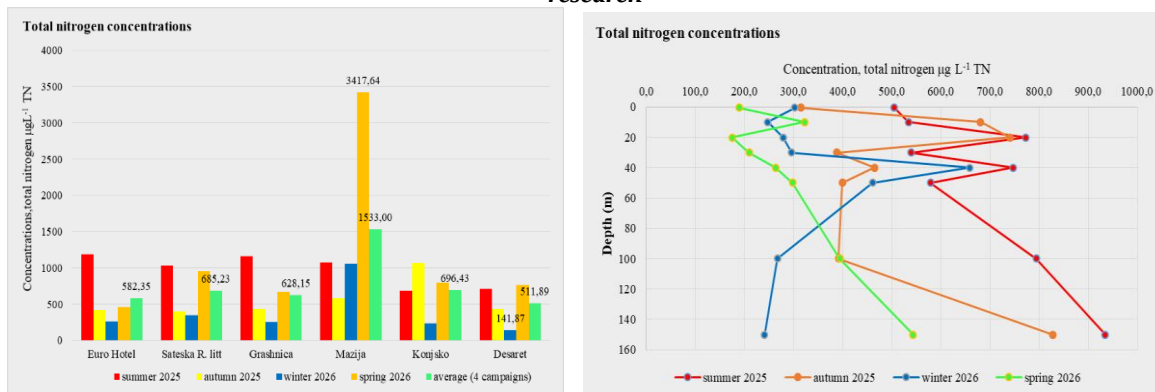
Overall, the present results indicate localized organic enrichment in the littoral zone, particularly at sites exposed to riverine and anthropogenic influences, whereas the pelagic zone shows lower and more stable

organic matter and BODs values. Together with the favourable dissolved oxygen regime, these findings are consistent with the generally oligotrophic character of Lake Ohrid, while confirming that certain littoral areas remain more sensitive to external organic loading.

3.3 Total nitrogen and total phosphorus are key nutrients regulating primary production in freshwater ecosystems and are therefore important indicators of water quality and trophic status. Increased nutrient loading may stimulate phytoplankton and macrophyte production, promote organic matter accumulation and, through subsequent microbial decomposition, increase oxygen consumption. Although phosphorus often plays a major role in freshwater eutrophication, both TN and TP contribute to phytoplankton development depending on trophic state and environmental conditions. A large-scale analysis of 1,382 lakes confirmed significant relationships between both nutrients and chlorophyll *a*, with phosphorus having a particularly important role under oligo-mesotrophic and eutrophic conditions (Liang et al., 2020).

The seasonal and average values (from the four seasonal campaigns for littoral zone) of total nitrogen (TN) concentrations, in the littoral and pelagic zones of Lake Ohrid are presented in Figure 4.

Figure 4. Total nitrogen concentrations at littoral and pelagic zones of Lake Ohrid - Source: Authors' research



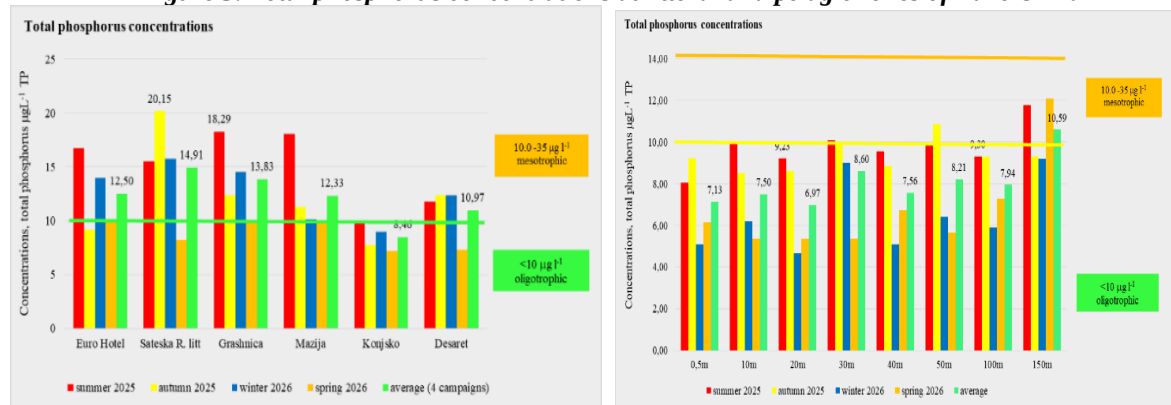
Source: Author's research

The littoral zone showed pronounced spatial and seasonal variability, with mean concentrations ranging from 511.89 $\mu\text{g L}^{-1}$ at Desaret to 1533.00 $\mu\text{g L}^{-1}$ TN at Mazija. The highest individual concentration (3417.64 $\mu\text{g L}^{-1}$ TN) was recorded at Mazija during spring 2026. The marked variability among littoral sites suggests localized nutrient inputs associated with tributaries, municipal wastewater, agricultural runoff, settlements and tourism.

In the pelagic zone, TN concentrations were generally lower and less variable, with mean values ranging from 327.65 $\mu\text{g L}^{-1}$ TN at 0.5 m to 636.62 $\mu\text{g L}^{-1}$ TN at 150 m. The highest individual value (934.51 $\mu\text{g L}^{-1}$ TN) was recorded at 150 m during summer 2025. Although no consistent increase with depth was observed, relatively higher TN concentrations in deeper layers may reflect nutrient regeneration during decomposition and mineralization of sinking organic matter, combined with reduced vertical exchange during stratification.

The seasonal and average values (from the four seasonal campaigns for littoral zone) of total phosphorus (TP) concentrations, in the littoral and pelagic zones of Lake Ohrid are presented in Figure 5.

Figure 5. Total phosphorus concentrations at littoral and pelagic zones of Lake Ohrid



Source: Author's research

Littoral TP showed clear spatial and seasonal variability, with mean concentrations ranging from 8.40 $\mu\text{g L}^{-1}$ TP at Konjsko to 14.91 $\mu\text{g L}^{-1}$ TP at the Sateska River littoral site. The highest individual concentration (20.15 $\mu\text{g L}^{-1}$) was recorded at Sateska during autumn 2025, while relatively elevated concentrations were also observed at Grashnica and Mazija. Based on the trophic ranges applied in this study, Konjsko remained within the oligotrophic range on average, whereas several other littoral sites showed values within the lower mesotrophic range. These results indicate localized phosphorus enrichment rather than a general shift of Lake Ohrid toward mesotrophic conditions. This interpretation agrees with previous investigations of Lake Ohrid. Schneider et al. (2014), studying 30 nearshore sites, found that TP concentrations in littoral waters were generally low, although macrophytes, benthic algae and macroinvertebrates showed biological responses to nutrient enrichment. Their results indicate that littoral communities may respond to local nutrient loading before corresponding changes become clearly detectable in pelagic water chemistry. In the pelagic zone, TP concentrations were more uniform and generally lower, with mean values ranging from 6.97 $\mu\text{g L}^{-1}$ TP at 20 m to 10.59 $\mu\text{g L}^{-1}$ TP at 150 m. Most of the water column therefore remained within the oligotrophic range, while a slight increase (above 10 $\mu\text{g L}^{-1}$ TP) was observed in the deepest investigated layer. Comparable vertical nutrient patterns have been reported in the deep oligotrophic Lake Traunsee, where seasonal stratification and biological nutrient utilization resulted in distinct vertical distributions of phosphorus fractions and TN:TP ratios (Teubner, 2003).

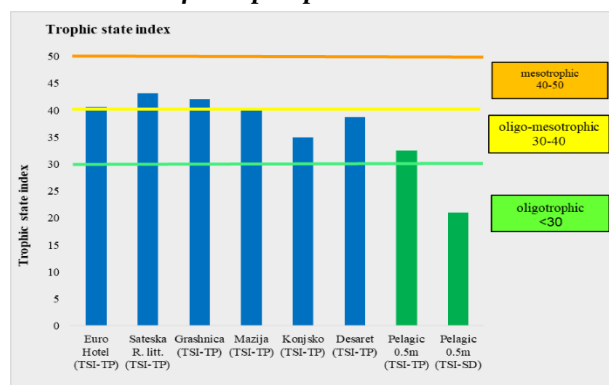
When considered together with dissolved oxygen, organic matter and BODs, the nutrient results reveal a consistent spatial pattern. The littoral zone showed greater variability and locally elevated TN, TP, organic matter and BODs, indicating greater sensitivity to catchment-derived and anthropogenic inputs. In contrast, the pelagic zone remained generally nutrient-poor, moderately loaded with organic matter and well oxygenated. Nevertheless, the somewhat higher TN and TP concentrations together with lower DO in the deepest investigated layer may indicate nutrient regeneration during mineralization of sinking organic material.

The potential long-term importance of phosphorus enrichment for Lake Ohrid has previously been demonstrated by Matzinger et al. (2006, 2007). Their studies showed that increasing phosphorus loading could enhance primary production and organic matter mineralization, resulting in greater oxygen consumption in deep waters; reduced vertical mixing could further amplify these effects.

Overall, the present results indicate that the pelagic zone largely retains the nutrient characteristics of an oligotrophic lake, whereas localized nutrient enrichment is evident at several littoral sites, particularly those exposed to riverine and anthropogenic influences. Continuous external inputs of nitrogen and phosphorus therefore represent an important long-term eutrophication risk.

3.4 Trophic state index -The trophic status of the investigated sites was additionally evaluated using Carlson's Trophic State Index (TSI) based on total phosphorus concentration and Secchi disk transparency. The calculated values are presented in Fig. 6. The lowest TSI value was recorded in the pelagic zone (TSI = 28), confirming its oligotrophic character. Higher values were obtained at several littoral sites, with maxima at Grashnica (TSI = 46) and the Sateska River littoral site (TSI = 47). Thus, the investigated littoral sites ranged from oligotrophic to transitional oligo-mesotrophic conditions, whereas the pelagic zone remained clearly oligotrophic. The TSI results support the patterns observed for TN, TP, organic matter and BODs. Sites with greater anthropogenic and riverine influence generally showed higher trophic-state values, while the open-water zone retained low nutrient concentrations, good transparency and favorable oxygen conditions. Importantly, the higher littoral TSI values represent localized trophic enrichment and should not be interpreted as evidence of a change in the trophic status of Lake Ohrid as a whole. This distinction is consistent with previous studies showing that eutrophication effects may first become evident in the littoral zone while pelagic phosphorus concentrations remain low (Schneider et al., 2014)

Figure 6. Trophic state index based of total phosphorus concentrations and secchi disk (SD) transparency



Source: Author's research

4. CONCLUSIONS

The combined assessment of dissolved oxygen, organic matter, BOD₅, TN, TP, and trophic-state indicators shows that the pelagic zone of Lake Ohrid largely retains the characteristics of an oligotrophic ecosystem. The water column remained well oxygenated, organic matter and BOD₅ were generally moderate, and TP was predominantly within the oligotrophic range. Slightly higher TN and TP concentrations in deeper water indicate the importance of continued monitoring of nutrient regeneration and deep-water oxygen dynamics.

The littoral zone showed greater spatial and seasonal variability, with localized increases in organic matter, BOD₅, TN, and TP. The strongest signals occurred at sites influenced by riverine or shoreline inputs, indicating that eutrophication pressure is currently more evident at a local littoral scale than in the pelagic zone. Importantly, the elevated loads observed during the study period did not result in pronounced oxygen depletion. Long-term monitoring should therefore focus on river mouths and other anthropogenically influenced littoral sites and should include TN and TP fractions, chlorophyll a, Secchi transparency, dissolved oxygen, BOD₅, and organic matter. Management priorities should include improved municipal wastewater collection and treatment, control of agricultural runoff and fertilizer losses, reduction of untreated discharges associated with settlements and tourism, and protection of littoral vegetation and wetland zones that contribute to nutrient retention. Preventing long-term phosphorus accumulation is particularly important because sustained enrichment may increase primary production, organic-matter mineralization, and deep-water oxygen demand.

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