Research Article

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Benthic invertebrates and associated fauna as bioindicators of water quality in the arroyo canal Candelaria, Southern Santa Fe Province, Argentina

Abstract

Benthic macroinvertebrates are widely used as bioindicators of ecological water quality due to their sensitivity to environmental disturbance and their central role in aquatic food webs. This study assessed the ecological condition of the Arroyo Canal Candelaria (ACC), a highly anthropized lowland stream in Southern Santa Fe Province, Argentina, using benthic macroinvertebrate assemblages and multiple biotic indices (IBMWP, IBF, IBPamp, ASPT), complemented by Shannon–Wiener diversity. A total of 6,584 individuals belonging to 22 families and several higher taxa were recorded. The upper and middle basin were dominated by tolerant groups (Oligochaeta, Nematoda, Chironomidae), exhibiting low richness and diversity, and consistently classified as "poor" or "very poor" by classical indices. In contrast, the lower basin showed increased richness, higher evenness, and the presence of insect groups indicative of improved habitat conditions. IBPamp, calibrated for Pampean streams, identified these sites as "good" to "very good," highlighting discrepancies with traditional indices and underscoring the importance of regionally adapted tools. Overall, the results reveal a clear longitudinal gradient of degradation–recovery, reflecting both the spatial distribution of pollution sources and the partial resilience of the system. The findings emphasize the need for targeted wastewater management in the upper and middle basin and demonstrate the value of combining classical and locally calibrated indices with community metrics to accurately assess ecological quality in lowland streams.

Keywords: benthic macroinvertebrates, bioindicators, water quality, lowland streams, pampean region

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Introduction

The study of lotic ecosystems and their ecological integrity is essential in the context of increasing environmental degradation and landscape fragmentation. Rivers and streams, in addition to supplying water and other key ecosystem services, function as ecological corridors that connect habitats, sustain biodiversity, and buffer anthropogenic impacts.[1,2] Within this framework, benthic macroinvertebrates have become a central component of biological monitoring in continental freshwater systems due to their sensitivity to multiple types of disturbance, their functional diversity, and their ecological role as essential components of aquatic food webs.[3,4]

Benthic macroinvertebrates—including aquatic insects, mollusks, annelids, crustaceans, and others—inhabit the bottom substrates of streams and rivers, occupying a wide range of microhabitats and hydrological conditions. Their life cycles span several weeks or months, with benthic stages long enough to reflect cumulative environmental changes.[5] Community structure responds clearly to factors such as organic load, current velocity, dissolved oxygen, sedimentation, and the presence of contaminants, making them reliable indicators of the ecological quality of aquatic environments.[6,7]

From an ecological perspective, streams are highly dynamic systems governed by longitudinal, lateral, and vertical connectivity.[8] This three-dimensional nature supports processes such as nutrient exchange, species movement, and the maintenance of critical habitats. However, in strongly anthropized landscapes—such as the agricultural regions of Central Argentina—streams are exposed to multiple pressures: loss of riparian vegetation, nutrient and agrochemical inputs, hydrological alteration, and fragmentation of the fluvial network.[9,10] These changes directly affect benthic communities, reducing their richness, functional diversity, and resilience.

Beyond their use in water quality assessment, macroinvertebrates play essential ecological functions. They process organic matter, promote nutrient recycling, and serve as a food resource for fish, amphibians, water birds, and other predators.[11] The loss of sensitive taxa or dominance of opportunistic species reflects not only pollution but also the structural and functional simplification of the ecosystem.

Methodologically, macroinvertebrate-based studies offer both logistical and scientific advantages. They are relatively cost-effective, allow the detection of cumulative impacts, and can be readily integrated into long-term monitoring programs. In addition, a robust body of literature and standardized protocols supports their application in both rural and urban contexts.[12,13]

In summary, the analysis of benthic macroinvertebrate communities integrates multiple ecological dimensions of fluvial ecosystems. Their study provides essential information to diagnose the ecological condition of a stream, understand its functioning, and guide management and restoration actions in bioindicators of water quality, we designed a methodological approach that combines field sampling, laboratory analysis, and the application of biotic indices. The following section describes the sampling, processing, and evaluation procedures used in this study.

Material and methods

Study area

The Arroyo Canal Candelaria (ACC) is located in the Southern sector of Santa Fe Province, within the Pampean ecoregion, in a landscape characterized by gentle hills, seasonally flooded depressions, and an intensive agricultural matrix. Despite its modest appearance, the ACC constitutes an ecologically and hydrologically relevant system at the regional scale: a watercourse of approximately 42 km that functions simultaneously as a stream, an anthropized drainage canal, and a wetland corridor (Figure 1).

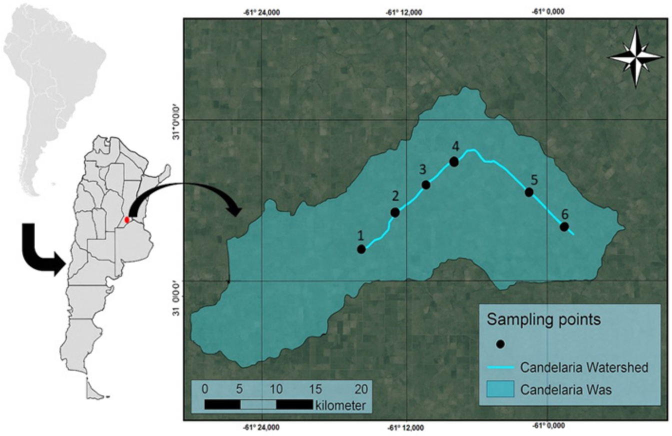

Figure 1 Basin of the Arroyo Canal Candelaria Stream, Southern Santa Fe province, Argentina.

The system originates near the town of Sanford, where the Perimetral Norte and Perimetral Sur canals converge, draining ancient paleochannels and low-lying areas of the landscape. From this point, the ACC flows southeastward until it discharges into the Arroyo Saladillo, thereby becoming part of the broader hydrological network of southern Santa Fe.

The ACC basin covers approximately 935 km² and exhibits the climatic characteristics typical of the Pampean plain: a warm-temperate and humid climate, with rainfall concentrated in spring and summer. Mean annual precipitation reaches 966.3 mm, generating pronounced hydrological pulses that influence lateral connectivity between the stream and its associated wetlands, the temporal expansion of inundated areas, and the availability of aquatic and semi-aquatic habitats. This hydrological regime—modulated by high summer evapotranspiration and extensive anthropogenic transformation of the territory—shapes key ecological processes within the system, including biotic structure, nutrient cycling, and water quality.

Working framework and applied methodology

Fieldwork

Following the methodological criteria proposed by Domínguez and Fernández (2009) and Lamas and Rimoldi (2019), sampling was conducted at six predetermined sites (points 1–6) along the Arroyo Canal Candelaria (ACC). At each site, a wadeable 10-m section of the streambed was surveyed, selecting various.

Benthic material was collected manually using a triangular hand net (25 × 34 × 34 cm) equipped with a 30-cm-long bag and a 250-µm mesh. The net was used specifically in areas exhibiting the greatest microhabitat diversity (presence of submerged vegetation, variable substrates, structural refugia) to obtain a representative sample of the system.

At each site, 125 cm³ of surface sediment were extracted, placed in a plastic container, and preserved with 10% formalin for subsequent laboratory processing. Each sample was carefully labeled with the following information: date, time, sampling campaign number, sampling site, and sample code.

At Site 6, the streambed was characterized by compact calcareous substrate (tosca) and deeper pools that hindered sweeping and dislodging of benthic material with the hand net. To minimize this bias, additional sweeps were performed along accessible margins and manual scraping of available substrate was carried out. Nonetheless, sampling efficiency was likely reduced at this site compared with others with softer substrates.

Laboratory procedures

Samples collected in the field were washed in the laboratory under running water using a 250-µm sieve to retain organisms. All material was transferred to white plastic trays, where macroscopic invertebrates visible to the naked eye were manually separated and preserved in 70% ethanol, in labeled vials corresponding to each sampling site.

To estimate the abundance of small benthic invertebrates, the remaining material—composed of fine particles and organisms not visible to the naked eye—was homogenized by gentle manual agitation to ensure random distribution. Subsamples of 2 cm³ were then taken using a standardized spoon. The material was stained with rose bengal dye to facilitate the visualization of organisms.[12–15] Preservation followed the same procedure applied to macroscopic invertebrates.

Subsamples were examined under a stereoscopic microscope, and all organisms present were counted and identified. Together with the macroscopic fraction, this method provided a representative estimate of the richness and relative abundance of the benthic assemblage in each sample.

Taxonomic identification and biological information were based on the following references: Hurlbert (1984), Brinkhurst and Marchese (1991), Lopretto and Tell (1995), Darrigran and Lagreca (2005), Angrisano and Sganga (2007), and Domínguez and Fernández (2009). Stereoscopic and compound microscopes were used to observe diagnostic morphological characters, and macro- and microphotographs were taken using a Samsung 8.1-megapixel camera. A laboratory spreadsheet was designed to record all fauna groups and their corresponding tolerance values for the biotic indices.[16–20]

Applied biotic indices

The ecological quality of the water was evaluated using four biotic indices based on the sensitivity of macroinvertebrates to organic pollution. These indices integrate taxonomic composition and ecological tolerance, providing an estimate of ecosystem condition:

I. **IBMWP (Iberian Biological Monitoring Working Party):** A score-based index that sums the values assigned to each macroinvertebrate family according to its sensitivity to pollution, with the most sensitive families receiving the highest scores.[21,22]

II. **IBF (Hilsenhoff Family Biotic Index):** A weighted-average index that incorporates the relative abundance of each taxon; higher values indicate greater levels of organic pollution.[23,24]

III. **IBPamp (Pampean Biological Index):** Developed specifically for lowland streams of the Pampean region, this index adjusts family tolerance scores to local ecological conditions, where the natural richness of Ephemeroptera, Plecoptera, and Trichoptera is low.[25,26]

IV. **ASPT (Average Score Per Taxon):** The average BMWP score per taxon present in a sample, providing a measure of the mean tolerance of the community, independent of taxon richness.[27]

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Species diversity

In addition to biotic indices, the Shannon–Wiener diversity index (H′) was calculated to analyze the structure of benthic macroinvertebrate communities at each sampling site. This index incorporates both taxon richness (number of distinct groups) and evenness (relative distribution of individuals among taxa), providing an integrated measure of local biodiversity.

The index was calculated as:

$$H' = -\sum_{i=1}^{S} \rho_i . In(\rho_i)$$

where p_i represents the proportion of individuals of taxon i relative to the total individuals in the sample, and S is the total number of taxa recorded.

Interpretation followed reference thresholds commonly used in limnological studies—non-absolute but useful for relative comparisons among sites:[28,29]

I. **H′ < 1:** very low diversity

II. **1 ≤ H′ < 2:** low diversity

III. **2 ≤ H′ < 3:** moderate diversity

IV. **H′ > 3:** high diversity

This approach complements pollution-sensitivity assessments by incorporating a structural perspective of the benthic community.

Results

A total of 6,584 aquatic invertebrate individuals were collected in the Arroyo Canal Candelaria (ACC), belonging to 22 families and several higher-level groups (Oligochaeta, Nematoda, Copepoda, Ostracoda, and Hirudinea). The community showed a marked dominance of a few groups: copepods (22.6%), oligochaetes (16.9%), nematodes (14.3%), and chironomids (14.2%), which together represented more than two-thirds of all organisms recorded. Additional groups with lower relative abundance included ostracods (8.3%) and hydrobiid snails (5.9%), indicating a community dominated by tolerant taxa, although with moderate lineage diversity (Figure 2).

Figure 2 Representative examples of invertebrates recorded in the Arroyo Canal Candelaria. References: **Top row:** (left) Aeglidae, (center) Chironomidae larva, (right) Hirudinea. **Middle row:** (left) Cladocera, (center) Ostracoda, (right) Copepoda with egg sacs. **Bottom row:** (left) Caenidae nymph, (center) Nematoda, (right) Oligochaeta.

Total abundance varied seasonally, with a maximum during winter (1,982 individuals, ~30% of the total), followed by similar values in summer (1,582; 24%), spring (1,544; 23.4%), and autumn (1,476; 22.4%). However, these differences were not statistically significant (ANOVA: $F = 0.195$; $p = 0.898$).

Family richness varied notably across the stream, ranging from 3 to 11 families per sample, with site means between 4 and nearly 7 families. Diversity, measured using the Shannon–Wiener index, also showed strong variability: the lowest values occurred in the upper basin, where communities were species-poor and dominated by a few groups, whereas the highest values were recorded in the lower basin, where organismal distribution was more even. Numerically, Shannon values ranged from 0.47 to 2.23 (equivalent to 0.68–3.22 bits in log base 2), confirming pronounced spatial heterogeneity. Upstream, the system was characterized by impoverished biota strongly influenced by pollution, whereas downstream segments showed signs of increased ecological complexity and higher taxonomic richness. Based on the abundance and occurrence of macroinvertebrates at each sampling point, several commonly used biotic indices were calculated to evaluate ecological water quality.

BMWP/IBMWP (Biological Monitoring Working Party)

The BMWP assigns scores to macroinvertebrate families according to their sensitivity to pollution, with sensitive families receiving high scores (up to 10) and tolerant ones receiving low scores. In the ACC, values were generally low, reflecting a scarcity of sensitive families.

In the upper basin (Sites 1 and 2), IBMWP values were close to 45, placing these sites in the "Poor" quality category (36–60). In the middle basin, values ranged from 49 to 60: Site 3 reached the upper limit of this category (~60), just below the threshold for "Moderate," while Site 4 remained around 50. Site 6 showed a similar pattern (~54), also classified as "Poor."

Only Site 5 showed a marked increase, with a BMWP of ~70, corresponding to "Moderate" quality (61–100). Overall, IBMWP indicated severe degradation from the upper to the middle basin, with slight recovery in the lower basin, particularly at Site 5. The absence or rarity of highly sensitive families—such as Plecoptera, Trichoptera, or diverse Ephemeroptera—strongly reduced scores throughout the system.

Note: Lower richness and relative abundance at Site 6 should be interpreted with caution; compact substrate (tosca) and deeper pools likely reduced sampling efficiency, potentially underestimating uncommon taxa or hard-to-access microhabitats, thereby affecting indices dependent on the detection of sensitive groups.

IBF (Hilsenhoff Family Biotic Index)

Unlike BMWP, IBF increases with pollution: values near 10 indicate very poor quality. In the ACC, high values confirmed the dominance of tolerant organisms. Site 1 (upper basin) showed an IBF of ~9.5, corresponding to "extremely poor" quality (≥7.26), reflecting severe organic pollution. Sites 2, 3, 4, and 5 ranged between 7.2 and 7.6, with no major differences among them, all within the categories of "Very poor" to "Extremely poor." Site 6 showed ~7.2, at the lower boundary of these categories. Community composition supported these patterns. At Site 1, over 90% of individuals were nematodes, oligochaetes, and tolerant dipteran larvae, explaining the extreme index value. Although additional aquatic insects occurred at other sites, resistant families (e.g., Chironomidae) remained dominant, maintaining values indicative of substantial organic pollution.

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IBPamp (Pampean Biological Index)

Designed for lowland Pampean streams, the IBPamp adjusts evaluations to local ecological conditions. Values obtained in the ACC showed a distinct pattern compared with classical indices, with more favorable ratings in the lower basin.

Site 5 reached ~10 ("Very Good"), while Site 6 scored ~8 ("Good"). By contrast, Sites 1–4 (upper and middle basin) yielded values between 6 and 7 ("Fair"). No site fell into the "Poor" or "Very Poor" categories.

This is expected for IBPamp: in Pampean lowland systems, the presence of families with intermediate tolerance increases scores. Thus, at Site 1, despite the impoverished fauna of the upper basin, the presence of Lestidae (Odonata)—absent in heavily polluted sites—prevented classification as "Poor." In the middle basin (Sites 3 and 4), the presence of Baetidae and Caenidae improved scores despite continued dominance of tolerant families.

The clearest divergence occurred in the lower basin: Site 5, rated "Poor" to "Very Poor" by BMWP and IBF, was classified as "Very Good" by IBPamp due to the presence of Hydroptilidae (Trichoptera) and odonates, and the relative scarcity of highly tolerant groups. This contrast is consistent with studies showing that traditional indices underestimate quality in Pampean streams, whereas IBPamp—calibrated for these systems—produces ratings more consistent with local ecological conditions.

ASPT (Average Score Per Taxon)

ASPT represents the average BMWP score per family present at each site, evaluating the mean sensitivity of the community. In the ACC, values were uniformly low (3.0–3.3), never exceeding 3.5. This reflects a community composed primarily of taxa with low to intermediate sensitivity. Even at Site 5—the site with the highest BMWP—the ASPT was only ~3.3, suggesting that although richness increased, many added families were of intermediate tolerance. ASPT values <4 are typically associated with environmentally stressed or degraded conditions, reinforcing the conclusion that the stream is structurally dominated by tolerant communities—a common pattern in systems with persistent organic loads and diffuse eutrophication.

Comparison of Indices and Overall Synthesis

Using multiple indices allowed the identification of a longitudinal gradient of improvement toward the downstream section, although the magnitude of this trend varied among indices:

I. **Site 1 (Upper basin):** All indices identified it as the most degraded site. IBMWP (~45) and IBF (~9.5) placed it in the worst quality classes ("Very Poor/Extremely Poor"), while ASPT (~3.2) confirmed dominance of tolerant families. IBPamp classified it as "Fair," but assigned one of the lowest values in the dataset.

II. **Sites 2, 3, and 4 (urbanized Middle basin):** These sites showed consistently poor conditions. IBMWP remained between 45–60 ("Poor"), and IBF between 7.3–7.6 ("Very Poor"). IBPamp values of 6–7 ("Fair") indicated slight improvement compared with the upper basin. Although additional aquatic insects appeared (Baetidae, Caenidae, Odonata), resistant families (e.g., Chironomidae, Oligochaeta) continued to dominate.

III. **Sites 5 and 6 (Lower basin, near the confluence with the Saladillo):** These sites represented the zone of greatest recovery. Site 5 showed the highest BMWP (~70, "Moderate") and a very high IBPamp (~10, "Very Good"). However, its IBF (~7.3) still reflected dominance of tolerant organisms. Site 6, with IBPamp ~8 ("Good") and IBF ~7.2 (the lowest in the series, though still "Very Poor"), suggested slight reduction of organic load. Its BMWP (~54) remained in the "Poor" category. Sampling difficulties at this site may have reduced detection of sensitive taxa.

Discussion

The integrated analysis of the macroinvertebrate community and biotic indices reveals a clear pattern of environmental degradation in the upper and middle basin of the Arroyo Canal Candelaria (ACC), with relative recovery in the lower basin. This longitudinal gradient is consistent with the spatial distribution of the main sources of impact.

The dominance of tolerant groups such as oligochaetes, nematodes, and chironomid larvae—particularly red "bloodworm" forms—in Sites 1–4 provides unequivocal evidence of organic pollution and hypoxic conditions, a pattern widely documented in urban and peri-urban streams of the Pampean region.[25,26] These organisms thrive in organic-rich environments where more sensitive taxa are absent. The low richness and Shannon–Wiener diversity observed in these reaches confirm this scenario: communities are impoverished and heavily dominated by a few resistant taxa. The scarce presence of aquatic insects with intermediate tolerance—such as Baetidae or Caenidae—in the middle basin suggests the existence of isolated microhabitats capable of sustaining them, although insufficient to reverse the overall degraded condition.

In contrast, the lower basin (Sites 5 and 6) showed a more diverse assemblage, including odonates, trichopterans, and even native decapods (Aeglidae), groups that require better oxygenation and greater habitat heterogeneity. This higher diversity and evenness, reflected in the elevated Shannon–Wiener values, provides complementary evidence to the biotic indices. While classical indices (BMWP, IBF, ASPT) still categorize these sites as "poor" in absolute terms, the IBPamp—calibrated for lowland Pampean streams—recognized the greater relative abundance of aquatic insects and assigned ratings of "good" to "very good" in the lower basin. This contrast supports previous findings emphasizing the need for indices adapted to the local biogeographical context to avoid overly pessimistic diagnoses in systems naturally poor in Ephemeroptera, Plecoptera, and Trichoptera.[10–30]

A particularly notable aspect is the discrepancy between BMWP/IBF and IBPamp ratings in the lower basin. Whereas the former penalize the absence of highly sensitive families, the latter interprets the presence of intermediate-tolerance groups as indicative of acceptable conditions. This reflects the differing conceptual foundations of the indices: classical indices were designed for European rivers with high EPT diversity, whereas adapted indices (IBPamp) acknowledge the naturally low representation of these orders in the Pampas. Consequently, a single site may be classified as "fair" or "good" by one index and "poor" by another, reinforcing the importance of comparative interpretation.

The improvement detected at Site 5 and the subsequent decline at Site 6 may be partly attributable to habitat-related differences in detectability. At Site 6, compact calcareous substrate and greater depth hindered the removal and retention of material with the hand net, increasing the likelihood of under-sampling cryptic microhabitats and less abundant taxa. This bias tends to depress observed richness and overrepresent tolerant groups, shifting index values downward. Thus, the apparent "decline" at Site 6 likely reflects both incomplete ecological recovery and reduced sampling efficiency due to bed conditions.

Despite methodological discrepancies, all indices and diversity metrics converge on two key points:

I. **Site 1 is the most impacted section**, with indicators of extremely poor biological quality attributable to anthropogenic inputs, dominance of resistant taxa, and low diversity.

II. **The Lower basin shows signs of self-purification and greater community heterogeneity,** demonstrating the stream's capacity for partial ecological recovery downstream, although not to pristine levels.

This longitudinal deterioration–recovery pattern has been documented in many regional watercourses subjected to urban and industrial pressures, where lower reaches may benefit from dilution and sedimentation processes but still retain strong signals of biological stress.[31,32]

An unexpected finding was the peak in abundance recorded during winter (~30% of all individuals), a pattern that contrasts with expectations for temperate systems, which typically show maxima in spring–summer. However, statistical analyses revealed no significant seasonal differences, indicating that the observed variations may reflect local fluctuations or stochasticity rather than marked seasonality. One possible explanation is the presence of continuous effluent discharges providing a stable year-round supply of nutrients and organic matter, favoring opportunistic taxa regardless of season. Additionally, local hydrological factors—such as lower discharge and increased sediment retention in winter—may enhance habitat availability for tolerant groups. Altogether, these results suggest that in highly anthropized systems, local factors may override climatic seasonality in structuring benthic communities.

From a management perspective, the results highlight the urgency of addressing point-source pollution in the upper and middle basin. The partial recovery observed near the downstream confluence indicates that the system retains resilience, but also that current pressures exceed its natural self-purification capacity. The control and treatment of domestic and industrial effluents are essential to improve ecological quality throughout the system allowed the characterization of the Arroyo Canal Candelaria (ACC) as a highly impacted system, with communities dominated by resistant organisms and low diversity, yet showing a positive trend toward recovery in the lower basin. This duality reflects both the methodological limitations of individual indices and the complex interaction between anthropogenic pressures and ecosystem response. The findings underscore the need for monitoring programs employing locally calibrated tools and for management strategies aimed at reducing organic loads in the upper and middle basin as a necessary condition to ensure the resilience and ecological functioning of the system.

Conclusion

This study demonstrates that the Arroyo Canal Candelaria (ACC) is a highly impacted lowland stream, with clear evidence of severe organic pollution and community impoverishment in its upper and middle reaches. The dominance of tolerant taxa, low richness, and reduced diversity in these sections reflect the influence of multiple anthropogenic pressures, particularly untreated domestic and industrial effluents. Despite these conditions, the lower basin exhibited signs of partial ecological recovery, with increased richness of aquatic insects, greater habitat heterogeneity, and improved biotic index scores—especially when evaluated with IBPamp, which is calibrated for Pampean lowland environments. The longitudinal gradient of deterioration followed by downstream recovery highlights both the resilience of the system and the limitations of its natural self-purification capacity. The discrepancies among biotic indices also emphasize the importance of using locally adapted tools when assessing ecological quality in biogeographically unique systems. Overall, our findings underscore the urgent need for effective wastewater management in the upper and middle basin and demonstrate the value of integrating classical and region-specific indices with community-level metrics. Such an approach provides a more accurate diagnosis of ecological condition and supports evidence-based management aimed at restoring the ecological functioning and long-term resilience of the ACC.

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None.

Conflicts of interest

The author declares there is no conflict of interest.

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