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NORTHAM BOOYSENDAL PLATINUM /

THE BUTTONSHOPE CONSERVATION TRUST

AQUATIC BIODIVERSITY STUDY

compiled as part of

BIODIVERSITY MANAGEMENT PLAN (BMP) STUDY FOR THE PROPOSED DE BERG PRIVATE NATURE RESERVE (DBPNR): 2021/22

AUGUST 2022

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Clean Stream Biological Services

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ABBREVIATIONS

ASPT	Average Score Per Taxon
BMU	Biodiversity Managament Unit
CBA	Critical Biodiversity Area
DEEEP	Standard Direct Estimation of the Ecological Effect Potential
DWAF	Department of Water Affairs and Forestry
DWS	Department of Water and Sanitation
EC	Electrical conductivity
EI	Ecological Importance
ES	Ecological Sensitivity
FAII	Fish Assemblage Integrity Index
FRAI	Fish Response Assessment Index
GSM	Gravel-Sand-Mud biotope
HCR	Habitat Cover Ratings (fish)
IHAS	Integrated Habitat Assessment System (invertebrates)
IHI	Index of Habitat Integrity
NFEPA	National Freshwater Ecosystem Priority Area
MBCP	Mpumalanga Biodiversity Conservation Plan
MTPA	Mpumalanga Parks and Tourism Agency
PES	Present Ecological State
S	Stones biotope
SASS5	South African Scoring System, Version 5
TWQR	Target Water Quality Range
VEG	Vegetation biotope
IWUL / WUL	Integrated Water Use Licence

1. INTRODUCTION

Clean Stream Biological Services (CSBS) was appointed by Northam Platinum Booysendal Mine to compile a first-phase biodiversity management plan (BMP) for the total surface rights area (approximately12 638ha) of the mine during 2020/21 period. This first-phase study indicated that the Northam Booysendal surface rights area, and especially the proposed De Berg Private Nature Reserve (DBPNR) (farms De Berg 71/0, 71/2, Triangle 72/0 and Sterkfontein 52/3), contains many plant and animal species and habitats of special conservation concern. Due to the critical biodiversity conservation importance of the proposed De Berg Private Nature Reserve area and the requirements for the establishment of the area as a private nature reserve, follow-up biodiversity assessments were conducted during 2021 and 2022 that specifically focused on the DBPNR area. The specialist information gathered as part of all work performed in this area during phase one and two (2020 to 2022) will be used to compile a BMP specifically for the DBPNR. The primary aim of these studies were to provide supporting information that is required as part of the application process in the proclamation of this important biodiversity conservation area as a private nature reserve. An additional section (farm Goedehoop) was added to the proposed DBPNR study area during early 2022 and limited baseline surveys were also conducted in this area.

Due to the current importance (priority) of the DBPNR in terms of biodiversity conservation and the need to conserve the upper catchment of the Groot Dwars River to ensure continued good water quality and flow in this river system, it was recommended that more detailed aquatic fauna assessments should be conducted in this area as part of the current phase of biodiversity studies for the proposed DBPNR. The following aspects were therefore included in the DBPNR aquatic ecology study (2021/22):

- Perform further aquatic biomonitoring surveys (fish, SASS5, diatoms) at selected sites within the DBPNR to expand the spatial and temporal information regarding the aquatic fauna diversity and ecological status of this area (included limited baseline diatom analyses, not performed in the study area before).
- Conduct detailed fish assessments of the dams in DBPNR to confirm the absence of fish (especially to ensure that no alien fish is present).
- Conduct focussed studies at selected sites to start an inventory of aquatic macro-invertebrates on species level (previous studies done on family level) and establish if any species of conservation concern are present.
- Conduct limited surveys (site inspection) on the recently new section (farm Goedehoop) added to the proposed DBPNR study area during early 2022.

The current report aims to provide detail regarding the **aquatic biodiversity** (fish, macroinvertebrates and diatoms) information of the proposed DBPNR study area based on the available literature and focussed aquatic fauna surveys conducted in the study are from 2020 to 2022.

2. STUDY AREA

The proposed De Berg Private Nature Reserve study area is situated approximately 30km to the west of the town of Lydenburg (Mpumalanga) (Figure 1). The original study area covered an area of the approximately 1881ha (farms De Berg 71/2, 71, Triangle 72/0 and Sterkfontein 52/3) with an additional section (farm Goedehoop) added to the study area during 2022. The acquisition of Goedehoop farm was driven by the intention to join the DBPNR with the existing Verlorenvalei provincial (MTPA) nature reserve. The proposed DBPNR forms part of Northam Booysendal mine offset agreement and falls under the management of the Northam Booysendal Mine, overseen by the Buttonshope Trust. The DNPNR study area falls within an extremely rugged topography and is situated on the eastern boundary of the Sekhukhuneland Centre of plant endemism and the northern edge of the floristically rich Steenkampsberg (and the Verlorenvalei N.R.).

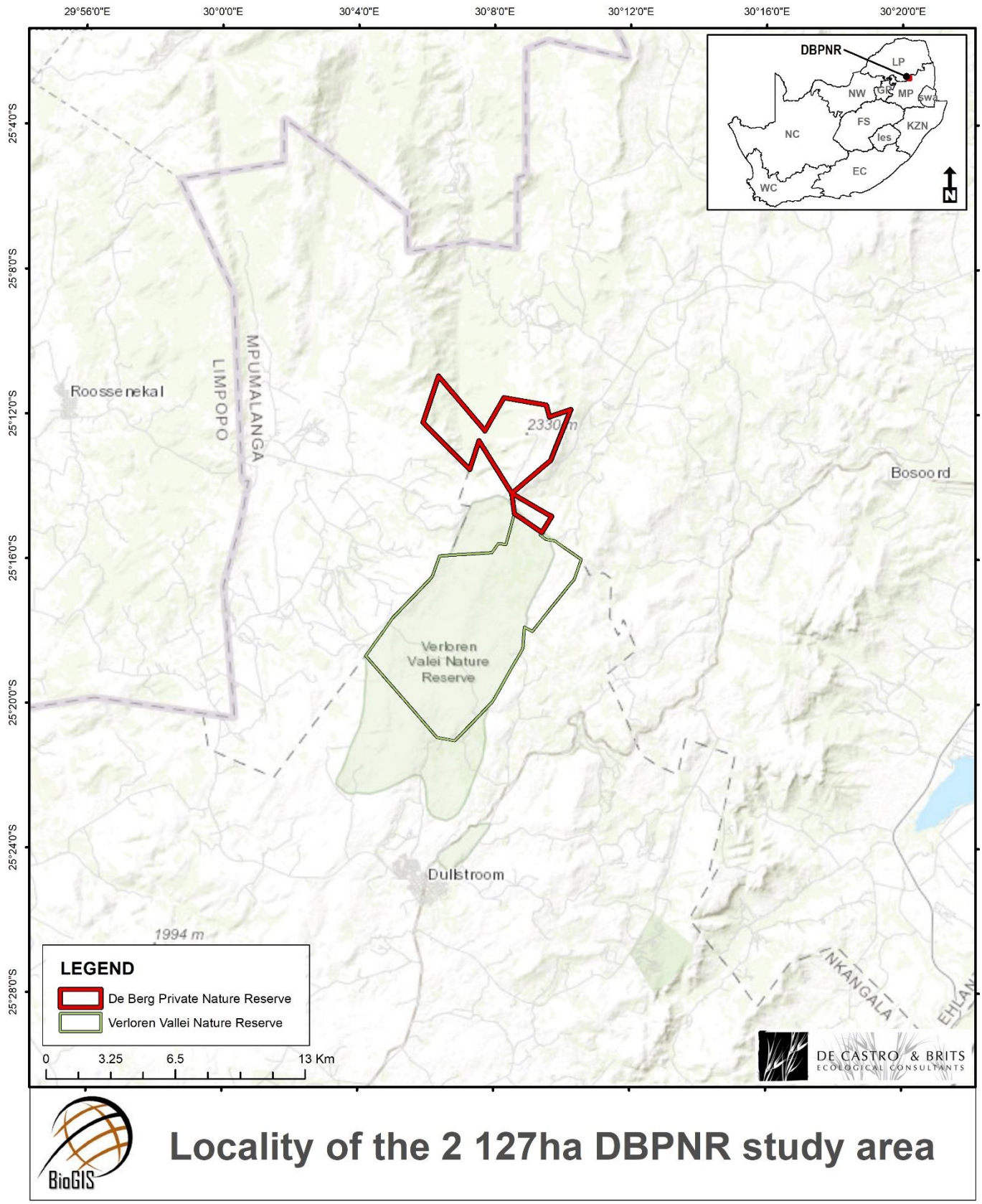

Figure 1: Locality map indicating DBPNR study area.

As part of a first-phase biodiversity study (Biodiversity Managament Plan) compiled in 2020/21 for the entire surface rights area of the Northam Booysendal mine, the area was divided into thirteen (13) biodiversity management units (BMU's) . The aim of defining biodiversity management units was to identify homogenous and discernible areas within Northam Booysendal, each with distinctive biodiversity composition, related aspects and management requirements. The broad-scale vegetation and land-use units provide a basis for the determination of different areas with homogenous characteristics, which in general also reflects discernible different faunal biotopes and are also visually identifiable within the study area. For these reasons, the vegetation/land-use units therefore formed the basis for the determination of the Biodiversity Management Units (BMU's). Eleven of the original 13 BMU's occur within the DBPNR study area. Within the DBPNR study area, the following three biodiversity management units are specifically relevant to the aquatic biodiversity study (current report) and refence is made to these units throughout this report (see Table 1 and Figure 1 for detail):

- **BMU 5:** Valley-bottom wetlands and seeps
- **BMU 7:** Mountain Streams
- **BMU 11**: Dams

Table 1: DBPNR Biodiversity Management Units (BMUs) of relevance to the aquatic biodiversity study.

Biodiversity Management Unit	Description
BMU 5: Valley-bottom wetlands and seeps	Includes the wetlands on hydromorphic soils of channelled and unchanneled valley-bottom wetlands and seeps. This unit also includes actively forming peat wetlands (mires) situated within valley-bottom wetlands and seeps on the Farm De Berg; sheetrock wetlands which comprise areas of exposed bedrock with patches of shallow to skeletal soils, located on mountain slopes or terraces and which experience temporary or seasonal surface flow and soil saturation. This unit comprising ca. 6.4% of the DBPNR. The wetlands comprising this unit also have exceptionally high functional value, form the source of the Groot Dwars River and provide crucial habitat for various species of fauna.
BMU 7: Mountain Streams	Includes the azonal riparian and instream zones of perennial and non-perennial mountain streams (mostly 1st, 2nd and 3rd order streams) which flow over both igneous, ultramafic rocks (mostly norite) and quartzites and arenite. This unit comprise ca. 2.5% of the study area. This unit has high functional value and provides crucial habitat for various species of fauna. All of the streams comprising this BMU form part of the catchments of the Groot Dwars River (including Everest tributary), Klip River and Potspruit.
BMU 11: Dams	This unit covers a total of 5 ha (or 0.2%) of the DBPNR and includes the artificial wetlands created by relatively small, earth-walled farm dams (on the farms De Berg and Goedehoop). According to the wetland classification system of (Ollis *et al*. 2013), all of the farm dams are 'in –channel' earth-walled dams.

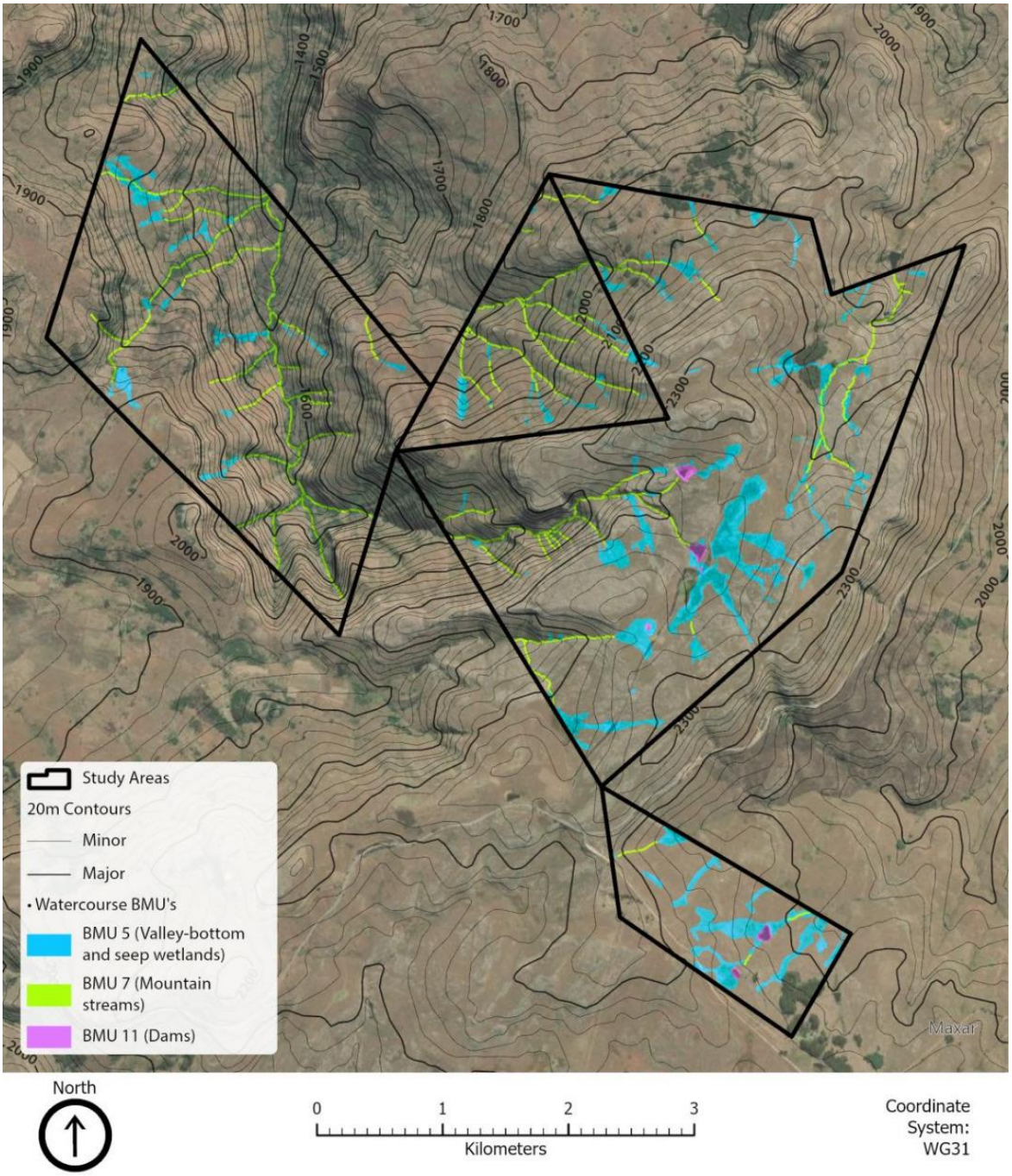

Figure 2: Map indicating aquatic Biodiversity Managament Units (BMUs) of the DBPNR study area.

In terms of aquatic ecosystems, the study area falls within the Olifants (B) Water Management Area (WMA) and specifically quaternary catchments B41G (Groot Dwars River and Everest tributary), B41C (Klip River) and B42F (Potspruit). The DBPNR study area is primarily drained by the upper (source streams) of the Groot Dwars River (sub-quaternary reach B41G-721), the most upper reaches of the Everest tributary that leaves the DBPNR to later flow into the Groot Dwars River outside of the current study area (before the inflow into Der Brochen Dam), and the upper Potspruit (on the farm Goedehoop) (Figure 3). The Groot Dwars Rivers (including the Everest tributary) confluence with the Klein Dwars River downstream of the study area to become the Dwars River that flows into the Steelpoort River (close to the town of Steelpoort), which is one of the primary tributaries of the Olifants River. The Klip River also flows into the Steelpoort River within the inundated section of the Goedehoop Dam. The Potspruit flows into the Waterval River (that includes the Buffelskloof Dam) and later joins the Spekboom River that also confluence with the Steelpoort River close to the town of Burgersfort. For the purpose of this aquatic fauna assessment emphasis is placed on the Groot Dwars River (SQ reach B41G-721) and its tributaries (Everest tributary and various other small unnamed

tributaries), the upper Klip River (B41G) as well as the upper reaches of the Potspruit (SQ reach B42F-812) (Figure 3).

Hydrogeomorphic Types

Five hydrogeomorphic types *sensu* Ollis *et al*. (2013) were identified in the Study Area as follows:

Wetlands	Streams	Artificial
• Hillslope Seeps	• Source Zones • Mountain Headwaters • Mountain Streams	• Earth Dams

Aquatic Biotopes

Nine aquatic biotopes *sensu* Ollis *et al*. (2013) were identified in the Study Area as follows:

- seeps
- springs
- bedrock
- pools
- runs
- riffles
- cascades
- waterfalls
- impoundments (Earth Dams)

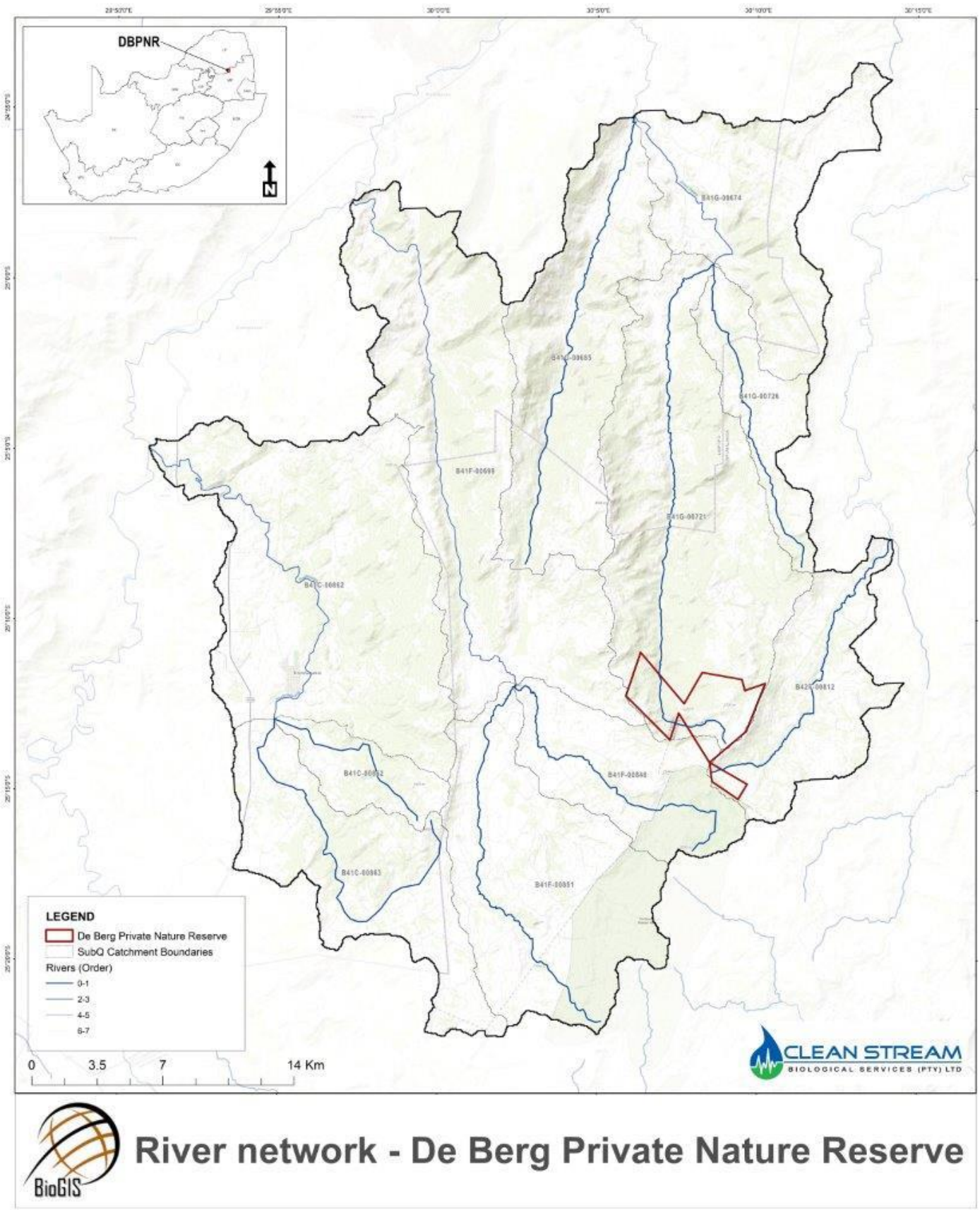

Figure 3: Map indicating rivers and catchments of concern in the DBPNR study area.

Limited aquatic surveys have been conducted on DBPNR as part of first-phase biodiversity project conducted in 2020/21. Representative sites were sampled in all the primary aquatic ecosystems within the DBPNR as part of the 2021/22 biodiversity study (Table 2, Figure 4).

Table 2: Description of the aquatic sampling sites sampled as part of the specialist surveys conducted within the proposed DBPNR study area.

River / Sub-reach	Sampling Site	Description	Latitude	Longitude
Groot Dwars River (B41G)	A1	Southern tributary upstream of de Berg Dam 1, comprising Seepage Wetlands, and Source Zone (**BMU5: Valley bottom wetlands and seeps**).	-25.221310°	30.149970°
	US-Dam1 / A2	Small stream (upper Groot Dwars River source) draining into De Berg Dam1 (at bridge crossing) / Eastern tributary upstream of de Berg Dam 1, comprising Seepage Wetlands, Source Zone and water impounded by Dam 1. (**BMU5: Valley bottom wetlands and seeps**).	-25.219093°	30.150664°
	De Berg_Dam1	Dam in upper reaches (source) of Groot Dwars River (at old farm house on De Berg). Altitude: 2229 m.a.s.l. **(BMU11: Dams)**	-25.220063°	30.149745°
	De Berg_Dam2	Small shallow dam in upper reaches (source) of Groot Dwars River (on De Berg). Altitude: 2230 masl. **(BMU11: Dams)**	-25.213944°	30.148555°
	GD_De Berg / A3	Upper reaches of Groot Dwars River directly downstream of De Berg_Dam1. Altitude: 2228 masl**. (BMU7: Mountain Stream)**	-25.218797°	30.148161°
	A4	Groot-Dwars River downstream of first waterfall, classified as a Mountain Headwater. **(BMU7: Mountain Stream)**	-25.215470°	30.143640°
	GD_SterkfonteinP3	Upper reaches of Groot Dwars River (on Sterkfontein Portion 3) Altitude: 1603 masl. **(BMU7: Mountain stream)**	-25.212070°	30.117870°
	GD N-trib	Upper reaches of a small (northern) tributary stream draining westwards into the Groot Dwars River (inside DBNR): **(BMU7: Mountain stream)**	-25.199307°	30.142752°
Everest Tributary (B41G)	Everest 1 / B1	Site in upper reaches of Everest Tributary (upstream of waterfall, mountain headwater) DBPNR. Altitude: 2170 masl. **(BMU7: Mountain stream)**	-25.206125°	30.161396°
	Everest 2	Site in upper reaches of Everest Tributary (downstream of waterfall, mountain headwater) DBPNR **(BMU7: Mountain stream)**	-25.205607°	30.163028°
	Everest 3	Site in upper reaches of Everest Tributary at boundary where it exits DBPNR **(BMU7: Mountain stream).**	-25.199954°	30.165214°
Klip River Catchment (B41C)	KR1 / C1	Upper reaches of an unnamed tributary of the Klip River (after confluence of two legs) on boundary of DBPNR (mountain headwater). **(BMU7: Mountain stream)**	-25.225939°	30.135030°
	KR2	Site in southern leg of upper reaches of an unnamed tributary of the Klip River inside DBPNR. **(BMU7: Mountain stream)**	-25.227377°	30.136228°

River / Sub-reach	Sampling Site	Description	Latitude	Longitude
Upper Potspruit (B42F-812)	GH Dam1	Dam 1 in the upper reaches of the Potspruit on the farm Goedehoop. **(BMU11: Dams)**	-25.249894°	30.152429°
	GH Dam2	Dam 1 in the upper reaches of the Potspruit on the farm Goedehoop. **(BMU11: Dams)**	-25.246917°	30.154842°
	PS1	Site in the upper reaches of the Potspruit downstream of GH Dam2 inside Goedehoop Farm section of proposed DBPNR. **(BMU7: Mountain stream, stretching into** (**BMU5: Valley bottom wetlands and seeps)**	-25.245823°	30.157139°

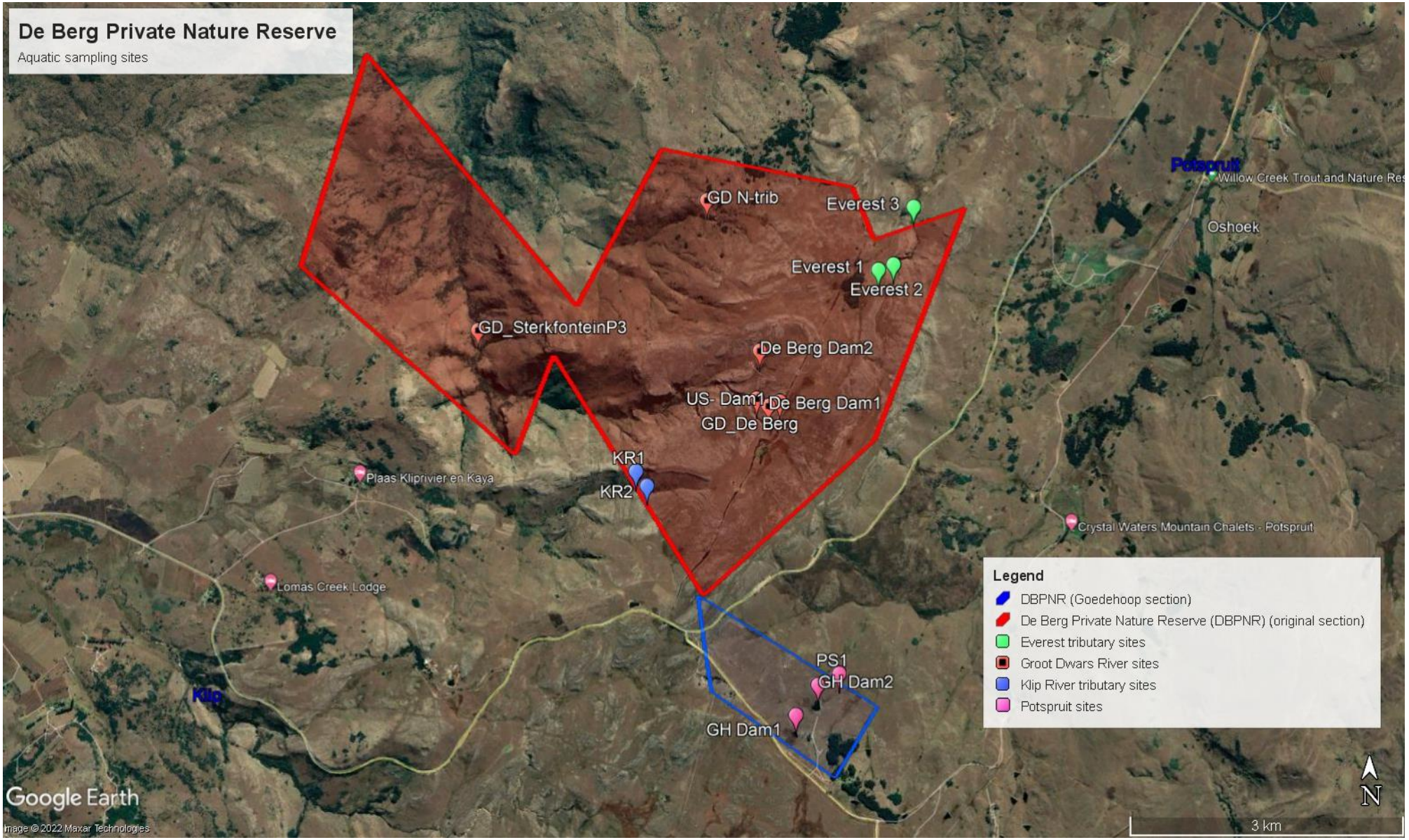

Figure 4: Aerial image (Google earth™) indicating primary aquatic ecosystems and location of various aquatic sampling sites within the DBPNR study area.

3. BACKGROUND INFORMATION

3.1 Legislative consideration

The National Water Act (Act No. 36 of 1998) (NWA) allows for the protection of water resources. The National Environmental Management Act (NEMA) (Act 107 of 1998) and the associated Regulations (No R. 982), as amended in December 2014, states that prior to any development taking place within a wetland or riparian area, an environmental authorisation process needs to be followed.

The Resource Quality Objectives for the Olifants River catchment (that includes the Northam Booysendal study area) were published in the Government Gazette in April 2016 (DWS 2016).

The National Environmental Management: Biodiversity Act No. 10 of 2004 addresses aspects such as protection of threatened ecosystems. The surrounding Steenkampsberge mountains function as natural drainage areas by intercepting rainfall and mist and channelling water to the receiving Groot Dwars River. Therefore, aspects of the Mountain Catchments Areas Act may also apply (although this is not a declared Mountain Catchment Area). The Act makes provision for the conservation and management of mountain catchments. Mpumalanga Nature Conservation Act (Act 10 of 1998) prohibits the creation of obstructions that may prevent the migration of fish and prohibits fishing with nets and pollution or disturbance of fish habitats. No person shall release fish (including exotic fish) into a watercourse without the required permit. The Act also applies to the propagation or removal of rare and protected species and management of alien plants and gives a list of “protected game”.

3.2 Present Ecological State (PES), Ecological Sensitivity (ES) and Ecological Importance (EI)

The Department of Water and Sanitation (DWS) conducted a desktop review and update of the present ecological status (PES), Ecological Sensitivity (ES) and Ecological Importance (EI) of all South African rivers (DWS, 2013). The desktop assessment was done on a sub-quaternary reach scale. The study also provided a description of the current land-use activities in each sub-quaternary catchment, as well as a desktop assessment of the present ecological state (PES), ecological importance[1] (EI) and ecological sensitivity[2] (ES) (DWS, 2014).

The two sub-quaternary reaches that was assessed in the study area include the Groot Dwars River (sub-quaternary reach B41G-721) and the Klein Dwars River (SQ reach B41G-685) (Figure 3). The total length of the Groot Dwars River SQ reach B41G-721 is 32km, with approximately 20km of this reach (upper section) falling within the Booysendal BMP study area (section from source to inflow into Der Brochen Dam). The total length of the Klein Dwars River SQ reach B41G-685 is 27km, with approximately 8km of this this river (upper reaches) running along the western boundary of the current Booysendal BMP study area.

Present Ecological State (PES)

The PES desktop assessment per sub-quaternary catchment considers the following criteria:

- Potential instream and riparian/wetland habitat continuity modification.
- Potential instream and riparian/wetland zone habitat modification
- Potential flow modification
- Potential physico-chemical modification

The desktop overall (instream and riparian zone) **PES** of the **Groot Dwars River** (sub-quaternary reach B41G-721 with a total length of 32.04 km) was classified as **Category C (moderately modified).** The PES

[1] Ecological importance (EI) of a river is an expression of its importance to the maintenance of ecological diversity and functioning on local and wider scales (Kleynhans 1999). Both abiotic and biotic components of the system are taken into consideration in the assessment of ecological importance (Kleynhans 1999).

[2] Ecological sensitivity (or fragility) (ES) refers to the system’s ability to resist disturbance and its capability to recover from disturbance once it has occurred (resilience) (Resh *et al*. 1988; Milner 1994; Kleynhans 1999).

of the small section of this Groot Dwars River sub-quaternary reach that falls within the DBPNR study area is significantly better due to a current low level of impacts and activities in these upper reaches, and can most probably be classified in a category A to B (largely natural to slightly modified). The desktop PES of the total **Potspruit** (SQ reach B41G-685 with a total length of 19.56km) was also classified in a **category C (Moderately modified). The very short section of this SQ reach that falls within the DBPNR study area (Goedehoop Farm)** is again estimated that it may be in a slightly better ecological state due to it being in the upper section with minimal impacts although it is currently impacted by flow modification (dams and potentially abstraction), erosion and livestock farming activities.

Table 3: Desktop present ecological status (PES), ecological importance (EI) and ecological sensitivity (ES) of the sub-quaternary reaches of concern.

METRIC	DESCRIPTION	Groot Dwars River (B41G-721: 32.04KM)	Potspruit (B42F-812: 19.56KM)
INSTREAM HABITAT CONTINUITY MODIFICATION	Modifications that indicate the potential that instream connectivity may have been changed from the reference.	LARGE	SMALL
RIP/WETLAND ZONE CONTINUITY MODIFICATION	Modifications that indicate the potential that riparian/wetland connectivity may have been changed from the reference	SMALL	SMALL
POTENTIAL INSTREAM HABITAT MODIFICATION ACTIVITIES.	Modifications that indicate the potential that instream habitats may have been changed from the reference. Includes consideration of the functioning of instream habitats and processes, as well as habitat for instream biota specifically.	MODERATE	MODERATE
RIPARIAN-WETLAND ZONE MODIFICATION	Modifications that indicate the potential that riparian/wetland zones may have been changed from the reference in terms of structure and composition that may influence these zones regarding functioning and processes occurring in the zones. Also refers to these zones as habitat for biota.	SMALL	SMALL
POTENTIAL FLOW MODIFICATION ACTIVITIES.	Modifications that indicate the potential that flow and flood regimes have been changed from the reference	MODERATE	MODERATE
POTENTIAL PHYSICO-CHEMICAL MODIFICATION ACTIVITIES	Activities that indicate the potential that physico-chemical conditions may have changed from the reference.	MODERATE	MODERATE
PRESENT ECOLOGICAL STATE		**C: Moderately modified**	**C: Moderately modified**

Ecological Importance

Kleynhans & Louw (2008) defined ecological importance of a river as its importance to maintain biological diversity and ecological functioning on a local and wider scale. The ecological sensitivity (or fragility) on the other hand refers to a river's ability to resist disturbance and its capability to recover from disturbances once it has occurred. Both abiotic and biotic components of the system are taken into consideration in the assessment of EI (Kleynhans 1999). The 2013 desktop assessment of the EI considered the following criteria:

- Fish and macroinvertebrate representivity and rarity.
- Riparian/Wetland/Instream vertebrates (excluding fish) importance.
- Riparian/Wetland natural vegetation in 500m zone.
- Riparian-Wetland vegetation importance.
- Habitat diversity.
- Habitat size (length).
- Instream and riparian-wetland zone migration link.
- Instream and riparian-wetland zone habitat integrity.

The desktop assessment concluded that the **ecological importance** of the Groot Dwars River (SQ reach B41G-721) and the Potspruit (SQ reach B41F-812) is **high** (Table 4).

Table 4: Desktop ecological importance (EI) metrics and overall rating of the sub-quaternary reaches of concern.

METRIC	DESCRIPTION	Groot Dwars River (B41G-721)	Potspruit (B42F-812)
FISH SPP/SQ	Number of species estimated per sub quaternary reach.	8	4
FISH REPRESENTIVITY PER SECONDARY CATCHMENT	Purpose is to identify the SQ with highest number of spp in the secondary catchment.	LOW	VERY LOW
FISH RARITY PER SECONDARY: CLASS	Rarity is based on the number of SQs a SPP occurs in relative to the total number of SQs in the Secondary Catchment.	MODERATE	VERY HIGH
ECOLOGICAL IMPORTANCE: RIPARIAN-WETLAND-INSTREAM VERTEBRATES (EX FISH) RATING	Rate the presence of ecologically important taxa in terms of biodiversity considerations, threatened, red listed, high spp richness etc.	LOW	HIGH
RIPARIAN-WETLAND NATURAL VEG RATING BASED ON % NATURAL VEG IN 500m	Based on the % natural vegetation (as land cover) within 500 m both sides of the stream and expressed as a rating from 0 to 5:	VERY HIGH	VERY HIGH
RIPARIAN-WETLAND NATURAL VEG IMPORTANCE BASED ON EXPERT RATING	Rate the presence of ecologically important taxa terms of biodiversity considerations, threatened, red listed, etc. high spp richness etc. Indicate important detail in comment cell.	LOW	LOW
INVERT TAXA/SQ	Estimated number of taxa per sub-quaternary reach	48.00	53.00
INVERTEBRATE REPRESENTIVITY PER SECONDARY CATCMENT	The purpose is to identify the SQ with highest number of taxa in the secondary catchment.	VERY HIGH	VERY HIGH
INVERTEBRATE RARITY PER SECONDARY:	Rarity is based on the number of SQs a taxon occurs in relative to the total number of SQs in the Secondary Catchment.	VERY HIGH	HIGH
ECOLOGICAL IMPORTANCE: RIPARIAN-WETLAND-INSTREAM VERTEBRATES (EX FISH) RATING	Rate the presence of ecologically important taxa terms of biodiversity considerations, threatened, red listed, high spp richness etc.	LOW	HIGH
HABITAT DIVERSITY	Assessment is relative to the SQ with highest length in the secondary catchment	VERY HIGH	MODERATE
HABITAT SIZE (LENGTH)	Assessment is relative to the SQ with highest length in the secondary catchment	HIGH	LOW
INSTREAM MIGRATION LINK	Assessment is directly related to the PES metric "instream habitat continuity"	MODERATE	VERY HIGH
RIPARIAN-WETLAND ZONE MIGRATION LINK	Assessment is directly related to the PES metric "riparian-wetland habitat continuity".	VERY HIGH	VERY HIGH

METRIC	DESCRIPTION	Groot Dwars River (B41G-721)	Potspruit (B42F-812)
RIPARIAN-WETLAND ZONE HABITAT INTEGRITY	Assessment is directly based on the PES metric "riparian-wetland zone habitat integrity".	VERY HIGH	VERY HIGH
INSTREAM HABITAT INTEGRITY	Assessment is directly based on the PES metric "instream habitat integrity".	HIGH	HIGH
OVERALL ECOLOGICAL IMPORTANCE		**HIGH**	**HIGH**

Ecological Sensitivity

Ecological sensitivity (or fragility) (ES) refers to the system's ability to resist disturbance and its capability to recover from disturbance once it has occurred (resilience) (Resh *et al*. 1988; Milner 1994; Kleynhans 1999). Both abiotic and biotic components of the system are taken into consideration in the assessment of ecological sensitivity (Kleynhans, 1999). The desktop assessment of the ES considered the following criteria:

- Fish and macroinvertebrates' intolerance level to physico-chemical modifications.
- Dependence on flow (velocity-depth) of fish and invertebrates.
- Riparian-Wetland-Instream vertebrates' (excl. fish) intolerance to water level/flow changes.
- Riparian-Wetland-Instream vegetation intolerance to water level/flow changes.
- Stream size sensitivity to modified flow/water level changes.

The desktop assessment concluded that the **ecological sensitivity** of the Groot Dwars River (SQ reach B41G-721) is **very high,** while the Potspruit (SQ reach B41F-812) is **high** (Table 5).

Table 5: Desktop ecological sensitivity (ES) metrics and overall rating of the sub-quaternary reaches of concern.

METRICS	DESCRIPTION	Groot Dwars River (B41G-721)	Potspruit (B42F-812)
FISH PHYSICO-CHEMICAL SENSITIVITY	Requirement of fish for unmodified water quality to survive and breed	VERY HIGH	HIGH
FISH NO-FLOW SENSITIVITY	Requirement of fish species for flow during different life stages.	VERY HIGH	HIGH
INVERT PHYSICO-CHEMICAL SENSITIVITY	Requirement of macroinvertebrates for unmodified water quality.	VERY HIGH	VERY HIGH
INVERTS VELOCITY SENSITIVITY	Requirement of macroinvertebrate taxa for flow.	VERY HIGH	VERY HIGH
RIPARIAN-WETLAND-INSTREAM VERTEBRATES (EX FISH) INTOLERANCE WATER LEVEL/FLOW CHANGES	In a SQ: the taxon with the highest rating (even if it uncommon), will represent the rating for this metric.	HIGH	HIGH
STREAM SIZE SENSITIVITY TO MODIFIED FLOW/WATER LEVEL CHANGES	Size of stream and morphology and geomorphic habitat units will determine sensitivity; consider degree of flow change that will elicit a particular level of response.	VERY HIGH	VERY HIGH
RIPARIAN-WETLAND VEG INTOLERANCE TO WATER LEVEL CHANGES	Sensitivity of riparian/wetland vegetation to change in water level.	LOW	LOW
OVERALL ECOLOGICAL SENSITIVITY		**VERY HIGH**	**HIGH**

3.3 National Freshwater Ecosystem Priority Area (NFEPA)

NFEPAs are rivers, together with their associated catchment, that are currently in a good to pristine state and are important in terms of maintaining threatened or near-threatened fish species. NFEPA Rivers should be maintained in a high level of biotic integrity in order to contribute to national biodiversity goals (Nel *et al.*, 2011) The river and its surrounding catchment, including tributaries, need to be managed in a way that maintains the good condition of the receiving river (A or B ecological category) (Nel *et al*., 2011).

The FEPA assessment indicated that the **Groot Dwars River** (SQ reach B41G-721) is **classified as a National Freshwater Ecosystem Priority Area** (Fish) (Figure 7). The fish species of conservation concern listed for this reach is *Opsaridium peringueyi*. This species is unlikely to occur within the Northam Booysendal study area (located upstream of the Der Brochen Dam) under present, and potentially also under reference conditions. This species is relatively abundant in the Steelpoort River and will also frequent the lower section of the Dwars River (outside the study area). The presence of another fish species of conservation concern, namely *Enteromius cf. motebensis* in the study area however confirms the importance of the Groot Dwars River as a priority area and hence further support the classification of this sub-quaternary reach as a NFEPA river. The Groot-Dwars river furthermore flows towards the Dwars River and Steelpoort River where *O. peringueyi* occurs. The NFEPA classification of the Groot Dwars River also indicated the presence of the following FEPA River Ecosystem types: Permanent/Seasonal - Eastern Bankenveld - Upper foothill and Lower foothill.

The **Potspruit** (SQ reach B41F-812) is **not classified as a NFEPA river** reach (fish). This sub-quaternary reach however includes FEPA wetland clusters, stating the following wetland ecosystem types to be present: Mesic Highveld Grassland Group 6_Channelled valley-bottom wetland, Mesic Highveld Grassland Group 6_Flat, Mesic Highveld Grassland Group 6_Seep.

The Mining and Biodiversity Guideline (DEA *et al.* (2013) recommends a buffer of 1 km from a FEPA River, while the Draft National Biodiversity Policy (2017) recommends an offset ration of 1:20 for FEPA rivers.

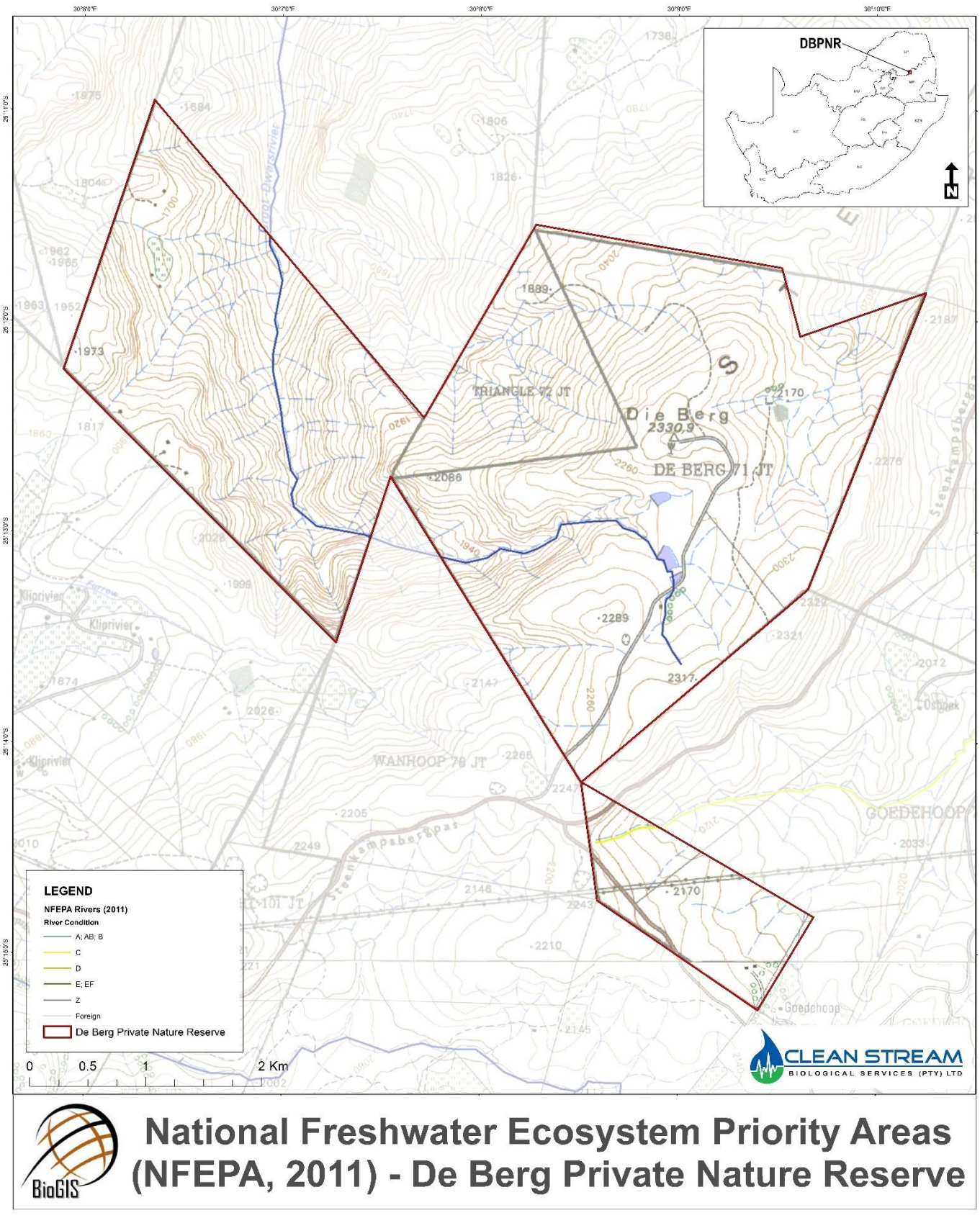

Figure 5: Position of DBPNR study area relative to NFEPAs.

3.4 Provincial Biodiversity Guidelines

Mpumalanga Biodiversity Sector Plan (MBSP)

The Mpumalanga Biodiversity Sector Plan (MBSP) is a spatial tool that forms part of a broader set of national biodiversity planning tools and initiatives that are provided for in national legislation and policy. It comprises a set of maps of biodiversity priority areas accompanied by contextual information and land-use guidelines that make the most recent and best quality biodiversity information available for use in land-use and development planning, environmental assessment and regulation, and natural resource management (MTPA, 2014).

The Groot-Dwars River (sub-quaternary reach B41G-00721) is considered by the Mpumalanga Biodiversity Conservation Plan to be **"Critical Biodiversity Areas"** (FEPA River), while the various tributaries draining these sub-catchments (Everest Tributary, etc.) are classified as "**Ecological Support Areas: Important sub-catchments"** (FEPA sub-catchments) (Figure 8).

The Mpumalanga Biodiversity Sector Plan classified the Study Area as follows:

- **ESA: Important Sub-catchment**. These are defined as "*Areas that are not essential for meeting biodiversity targets, but that play an important role in supporting the functioning of protected areas or CBAs and for delivering ecosystem services.*" The overall management objective for these areas is to "*minimise habitat and species loss and ensure ecosystem functionality through strategic landscape planning*" (MTPA 2011);

- **CBA: Rivers.** The Groot Dwars River was classified as a "*CBA: Rivers"*. *These are defined as "areas of high biodiversity value and need to be kept in a natural state, with no further loss of habitat or species*" (MTPA 2011);

- **CBA: Wetlands.** Wetlands in the southern portion of the Study Area were classified as a "*CBA: Wetlands"*. *These are defined as "areas of high biodiversity value and need to be kept in a natural state, with no further loss of habitat or species*" (MTPA 2011); and

- **ESA: Wetland Clusters.** The southern portion of the Study Area was classified as "*ESA: Wetland Clusters*". These are described as "*wetlands embedded within a largely natural landscape to allow for the migration of fauna and flora between wetlands"* (MTPA 2011).

Table 6: Summary of map categories shown in the freshwater/aquatic CBA map for Mpumalanga, and their meanings (MTPA, 2014).

Map category	Description	Subcategory	Description
Critical Biodiversity Areas (CBA)	All areas required to meet biodiversity pattern and process targets; Critically Endangered ecosystems, critical linkages (corridor pinch-points) to maintain connectivity; CBAs are areas of high biodiversity value that must be maintained in a natural state.	CBA: Rivers	Rivers, with a 100 m buffer, that need to be maintained in a good ecological condition in order to meet biodiversity targets for freshwater ecosystems. This category includes FEPA rivers and all FEPA free-flowing rivers. The FEPA rivers include those required to meet biodiversity targets for threatened fish species.
		CBA: Wetlands	Wetlands that are important for meeting biodiversity targets for freshwater ecosystems; the ecological condition of these wetlands needs to be maintained or improved, and their loss or deterioration must be avoided. This category includes FEPA wetlands.
		CBA: Aquatic Species	Areas considered critical for meeting the habitat requirements for selected aquatic invertebrate species (dragonflies, damselflies, crabs). These species are known to occur only at one or a few localities and are at high risk of extinction if their habitat is lost. Fish species are included under the CBA River category.
Ecological Support Areas (ESA)	Areas that are not essential for meeting targets, but that play an important role in supporting the functioning of	ESA: Wetland Clusters	Clusters of wetlands embedded within a largely natural landscape to allow for the migration of fauna and flora between wetlands.
		ESA: Wetlands	All non-FEPA wetlands. Although not classed as FEPAs, these wetlands support the hydrological functioning of rivers, water tables

Map category	Description	Subcategory	Description
	CBAs and that deliver important ecosystem services		and freshwater biodiversity, as well as providing a host of ecosystem services through the ecological infrastructure that they provide.
		ESA: Important Sub-catchments	Sub-catchments that either contain river FEPAs and/or Fish Support Areas.
		ESA: Fish Support Area	Sub-catchments that harbour fish populations of conservation concern, based on FEPA data augmented with regional data sets.
		ESA: Strategic Water Source Areas	High rainfall areas that produce 50% of Mpumalanga's runoff in only 10% of the surface area, thus supporting biodiversity and underpinning regional water security.
Other Natural Areas (ONA)	Areas that have not been identified as a priority in the current systematic biodiversity plan but retain most of their natural character and perform a range of biodiversity and ecological infrastructural functions.		
Heavily Modified Areas	Areas in which significant or complete loss of natural habitat and ecological function has taken place due to activities such as ploughing, hardening of surfaces, open-cast mining, cultivation and so on.	Heavily Modified	Heavily Modified: All areas currently modified to such an extent that any valuable biodiversity and ecological function has been lost.
		Heavily Modified: Dams	Artificial water bodies that have impacted on wetland or river ecosystems. These areas may still have a recharge effect on wetlands, groundwater and river systems and may support river- or water-dependent fauna and flora, such as water birds and wetland vegetation.

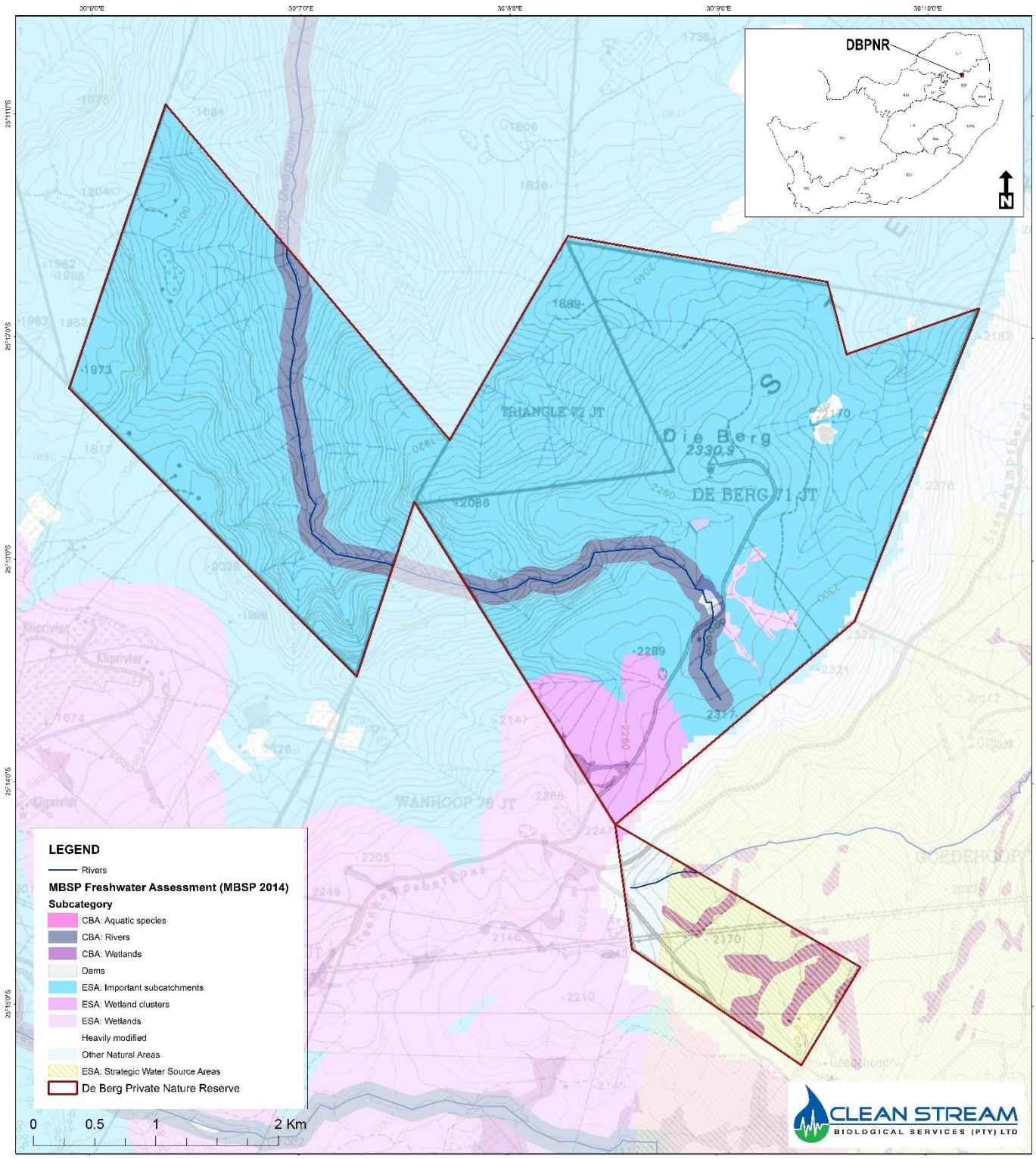

Figure 6: The location of DBPNR study area relative to the classification of aquatic biodiversity sub-catchments in the Mpumalanga Conservation Plan (2013).

3.5 Aquatic Biodiversity Sensitivity

The National Environmental Screening Tool indicated that the aquatic biodiversity sensitivity of the majority of the DBPNR study area was ***Very High*** (Figure 9). The reasons given for the very high classification were:

- Critical Biodiversity Areas (CBAs) for aquatic ecosystems
- wetland features; and
- freshwater priority areas.

A section of the study area (new Goedehoop farm section) falls with the ***low sensitivity*** zone (Figure 9).

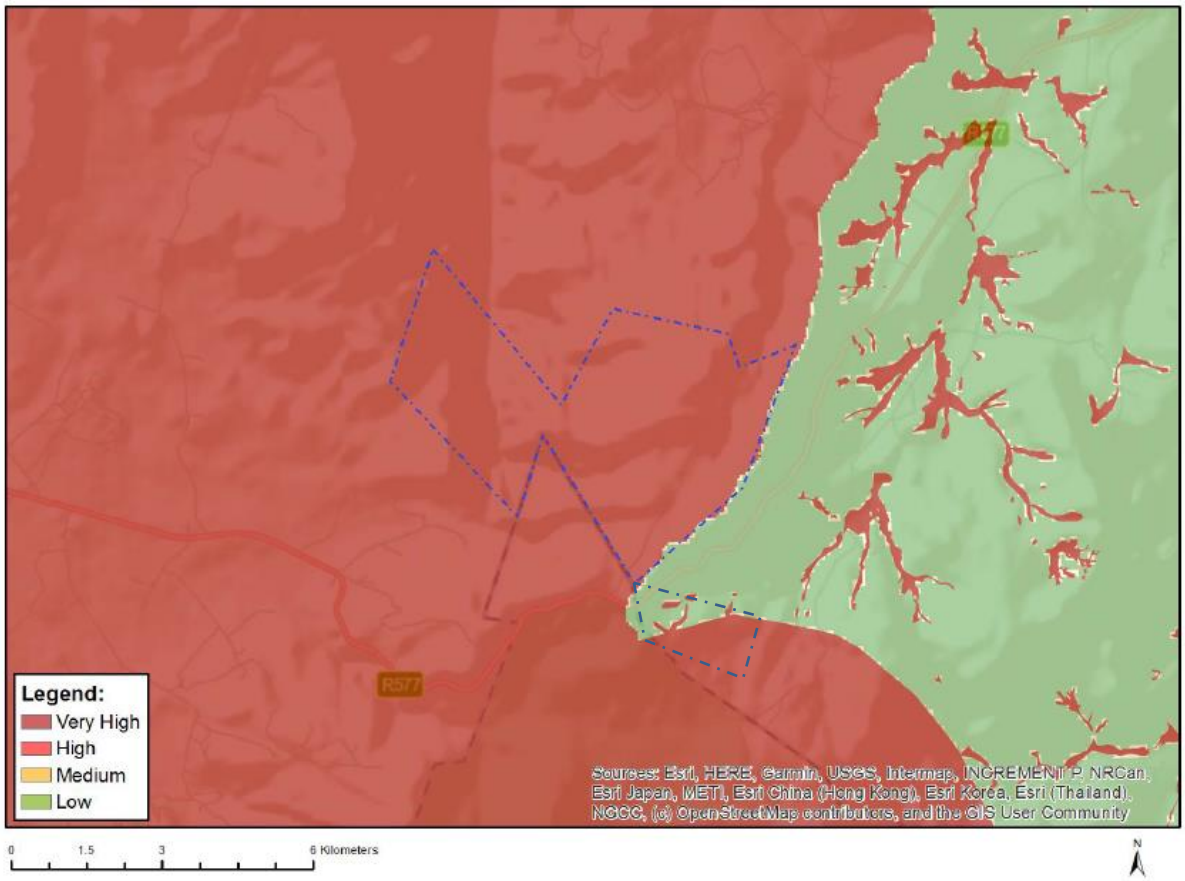

Figure 7: Aquatic Biodiversity Theme Sensitivity

[Source: Environmental Screening Tool (https://screening.environment.gov.za).]

4. DBPNR AQUATIC SURVEYS (2020-2022)

Aquatic sampling (fish and macroinvertebrates) was conducted at a few sites within the DBPNR area as part of the first-phase biodiversity study for the Northam Booysendal surface rights area (2020/21). It was recommended that more sites should be sampled to expand the spatial and temporal information regarding aquatic fauna within this area during the current study (2021/22). A more detailed aquatic macroinvertebrate study was also initiated and baseline diatom data was collected at selected sites. A more detailed fish survey was also conducted on the dams within the Groot Dwars River system within the DBPNR (on the farm De Berg) to verify the presence/absence of alien fish species within these dams previously stocked with alien Rainbow trout.

4.1 Physico-chemical habitat / water quality

Selected water quality variables were recorded on-site at the time of biological sampling to provide a broad indication of water quality conditions and to assist in the interpretation of biological results. Also refer to diatom section below that provides an indication of the biological water quality of selected sites.

The electrical conductivity measured in the **Groot Dwars River** system was very low (1.4 mS/m) in the most upper reaches of the Groot Dwars River at site GD-US Dam1 (BMU5: wetlands and seeps), confirming very low salinity prevailing in these upper reaches (Table 7). Salinity increased only slightly downstream of GD Dam1 (8.4 mS/m at site GD_De Berg) and remained very similar towards site GD_SterkfonteinP3, indicating overall low salinity and good water quality prevailing in the most Upper Groot Dwars River reach inside DBPNR. A similar scenario was also observed in the unnamed northern tributary of the Groot Dwars River (GD N-trib) within the DBPNR where an EC level of 8.6 mS/m was recoded (Table 7). The pH measured circum-neutral at all Groot Dwars River sites and also fell within guideline limits for fish health of 6.5 to 9 (DWAF 1996).

A similar scenario was observed in the **Everest tributary** with all sites reflecting very low salinity (1.5 to 6.5 mS/m) and circum-neutral pH levels falling within general guideline values (Table 7). A relatively low EC was also recorded in the **Klip River tributary** (site KR1) with pH values falling within general guideline limits (Table 7).

Table 7: On-site water quality variables measured at the time of sampling at the selected additional sampling sites (2020 to 2022).

Monitoring site	EC (mS/m)	pH	Water temp (ºC)	Turbidity (visual)	Flow (visual)	Time	Date
US-Dam1	1.4	7.9	17.9	Clear	Low	15:25	08/02/2022
GD_De Berg	8.4	7.8	n/a	Clear	Low	8:00	10/12/2020
GD_SterkfonteinP3	5.9	7.1	n/a	Clear	Moderate	11:00	09/12/2020
GD N-trib	8.6	8.8		Clear	Low	10:30	08/02/2022
Everest 1	1.5	7.3	21.4	Clear	Low	13:30	10/12/2020
Everest 2	1.5	9.0	20.7	Clear	Low	13:15	08/02/2022
Everest 3	6.5	7.8		Clear	Moderate	15:00	08/02/2022
KR1	9.9	7.9		Clear	Moderate	11:00	08/02/2022

4.2 Diatoms (Koekemoer, 2022)

Diatoms are of great ecological importance because of their role as primary producers, and they form the base of the aquatic food web. Diatoms have been shown to be reliable indicators of specific water quality problems such as organic pollution, eutrophication, acidification and metal pollution, as well as for general water quality. This information however also provide preliminary data on the diversity of diatom species in an area. It was therefore recommended that diatom analyses should be included at selected sites to gather baseline data and verify the biological water quality, as well as to describe the current diatom diversity of the aquatic ecosystems in the study area. Representative sites were sampled within the proposed DBPNR study area to gain some baseline diatom results of the area during the February 2022 aquatic survey.

While algal based bio-assessments in streams have been extensively researched worldwide and applied in regular riverine and lake monitoring programmes with great success, including South Africa, the application of diatoms as bio-indicators in wetland ecosystems are still under development. The scope of the current diatom assessment was to determine:

- If the water courses contained any endemic diatom species;
- What the biological water quality was;
- To identify and assess the main anthropogenic activities impacting the water courses.

The measure of relative abundance and species composition (i.e. assemblage patterns) to infer baseline water quality conditions at the water courses were applied. The objectives included:

- Characterisation of the diatom species and communities in terms of the water quality preference of dominant species.
- Presence of metal toxicity based on the presence of deformities.
- Identification of Key Performance Indicators/species in an attempt to determine the main impacts, if any, on the water courses.

A summary of the February 2022 diatom results is provided in Table 8 and the associated species list is provided in Addendum B (Koekemoer, 2022). Species contributing 5% or more to the total count were classified as dominant.

Table 8: Results of diatom analysis: Biological water quality

Site	No	SPI score	Class	Category	PTV (%)	Valve
US-Dam1	32	19	High quality	A	0	0
KR1	8	19.9	High quality	A	0	0
Everest2	11	19.1	High quality	A	0	0
Everest3	18	19.4	High quality	A	0.3	0
GD N-Trib	36	11.9	Moderate	C/D	0.5	0

SITE US-DAM1 (Groot Dwars River: BMU 5: Wetlands and seeps)

Taxonomic composition: A total of 33 diatom taxa (32 species and 1 variety) from 16 genera were identified in the epilithon of the stream. Seven taxa were determined to generic level. The genera *Eunotia* (9 taxa), *Achnanthidium* (5 taxa) and *Frustulia* (5 taxa) had the greatest number of species. Although no endemic species were observed, the dominant species from the above-mentioned genera were sensitive to water quality change with a preference for slightly acidic oligotrophic waters. The dominant species that included *Tabellaria flocculosa*, *Eunotia minor* and *Eunotia bilunaris,* are key indicator species of anthropogenically unimpacted sites with high biological water quality and reflected the near pristine condition and high biological water quality of the site (Table 8).

Biological water quality: During the February 2022 biodiversity assessment, US-Dam1 obtained a Specific Pollution sensitivity Index (SPI) score of 19, reflecting high biological water quality (Ecological Category A; Table 8). Nutrient levels, organic load and salinity concentration were low and further analysis of the various indices within OMNIDIA suggested pollution levels were slight. Overall, the diatom community consisted of

sensitive species with a preference for high water quality. Based on the diatom community composition no impact from anthropogenic activity was evident. No valve deformities were observed indicating that no metal toxicity was present.

SITE GD N-TRIB (Groot Dwars River tributary: BMU7: Mountain stream)

Taxonomic composition: A total of 36 diatom taxa (35 species and 1 variety) from 22 genera were identified in the epilithon of the stream. Three taxa were determined to generic level. The genera *Gomphonema* (6 taxa), and *Eunotia* (7 taxa) had the greatest number of species. Three endemic species were observed which occurred at low abundance and included *Achnanthes subsaxonica, Encyonema theronii* and *Eunotia hugenottarum*.

Biological water quality: During the February 2022 biodiversity assessment, Site GD N-Trib obtained a SPI score of 11.9, reflecting moderate biological water quality (Ecological Category C/D; Table 8). Nutrient levels, organic load and salinity concentration were low and further analysis of the various indices within OMNIDIA suggested pollution levels were slight. The species diversity was notably higher compared to the rest of the sites in the study area and the composition of the diatom community suggested a recent disturbance at the site associated with a recent flushing event. *Achnanthidium* and *Ulnaria* species were dominant and are associated with flushing events. This could be the reason for the high abundance of *Gomphonema lagenula* which is associated with increased sedimentation and has a preference for higher organic load and resulting in deteriorated water quality. However, sensitive *Eunotia* species along with sensitive endemic species were prolific, suggesting that the deteriorated water quality was associated with natural variability within the aquatic system and not anthropogenic impact. No valve deformities were observed indicating that no metal toxicity was present.

SITE EVEREST 2 (Everest tributary: BMU7: Mountain stream)

Taxonomic composition: A total of 11 diatom taxa (10 species and 1 variety) from 7 genera were identified in the epilithon of the stream. One taxon was determined to generic level. The genera *Achnanthes* (3 taxa), had the greatest number of species. Four endemic species were observed of which three were from the genus *Achnanthes - Achnanthes standeri*, *Achnanthes subsaxonica* and *Achnanthes subaffinis*. These three species dominated the diatom community by 78% and have a preference for well oxygenated, oligotrophic slightly acidic fresh waters (Taylor *et al*., 2007b). The other endemic, *Encyonema theronii* occurred at low abundance. is typical of oligotrophic waters with extremely low electrolyte content and sandstone bedrock (Taylor, *pers. comm*., 2022*).* An undescribed *Gomphonema* species, which was also dominant, was observed.

Biological water quality: During the February 2022 biodiversity assessment, Everest2 obtained a SPI score of 19.1, reflecting high biological water quality (Ecological Category A; Table 8). Nutrient levels, organic load and salinity concentration were low and further analysis of the various indices within OMNIDIA suggested pollution levels were slight. Overall, the diatom community was composed of sensitive species with a preference for high water quality, reflecting near pristine conditions. Based on the diatom community composition no impact from anthropogenic activity was evident. No valve deformities were observed indicating that no metal toxicity was present.

SITE EVEREST 3 (Everest tributary: BMU7: Mountain stream)

Taxonomic composition: A total of 18 diatom taxa (17 species and 1 variety) from 11 genera were identified in the epilithon of the stream. Six taxa were determined to generic level. The genera *Achnanthes* (3 taxa), and *Eunotia* (4 taxa) had the greatest number of species. Five endemic species were observed of which three were from the genus *Achnanthes - Achnanthes standeri*, *Achnanthes subsaxonica* and *Achnanthes subaffinis*. These three species dominated the diatom community by 58% and have a preference for well oxygenated, oligotrophic slightly acidic fresh waters (Taylor *et al*., 2007b). The other two endemics, *Encyonema theronii* and *Eunotia hugenottarum* occurred at moderate abundance and have a preference for high water quality (Taylor, *pers. comm*., 2022*). Nupela* species also occurred at high abundance and suggested that the water was acidic with low conductivity (Taylor and Cocquyt, 2016). The undescribed *Gomphonema* species observed at Site Everest2, was also present at Site Everest3.

Biological water quality: During the February 2022 biodiversity assessment, Everest2 obtained a SPI score of 19.4, reflecting high biological water quality (Ecological Category A; Table 8). Nutrient levels, organic load

and salinity concentration were low and further analysis of the various indices within OMNIDIA suggested pollution levels were slight. Overall, the diatom community reflected that sensitive species with a preference for high water quality were prolific, reflecting near pristine conditions. Based on the diatom community composition no impact from anthropogenic activity was evident. No valve deformities were observed indicating that no metal toxicity was present.

SITE KR1 (Klip River tributary: BMU7: Mountain stream)

Taxonomic composition: A total of 8 diatom taxa (8 species) from 7 genera were identified in the epilithon of the stream. Seven taxa were determined to generic level. The genera *Achnanthes* (1 taxon), *Encyonema* (1 taxon), and *Stenopterobia* (1 taxon) had the greatest number of species. Two endemic species were observed. These two species were dominant and included *Encyonema theronii* and *Achnanthes standeri* and made up 86% of the community. *Achnanthes standeri* is found in well oxygenated, oligotrophic slightly acidic fresh waters (Taylor *et al*., 2007b) while *Encyonema theronii* is typical of oligotrophic waters with extremely low electrolyte content and sandstone bedrock (Taylor, *pers. comm*., 2022). *Stenopterobia delicatissima* occurs in acidic, oligotrophic (sometimes dystrophic) upland waters with low to moderate electrolyte content (Taylor *et al*., 2007b) and an indicator of high quality water. The dominant species reflected the pristine condition and high biological water quality of the site (Table 8).

Biological water quality: During the February 2022 biodiversity assessment, KR1 obtained a SPI score of 19.9, reflecting high biological water quality (Ecological Category A; Table 8). Nutrient levels, organic load and salinity concentration were low and further analysis of the various indices within OMNIDIA suggested pollution levels were slight. Overall the diatom community reflected low species diversity with only sensitive species with a preference for high water quality present. Based on the diatom community composition no impact from anthropogenic activity was evident. No valve deformities were observed indicating that no metal toxicity was present.

4.3 Mosses and Algae (Palmer, 2022)

Mosses and algae comprise a significant component of primary production within aquatic ecosystems, not just on the seepage wetlands, but in all hydrogeomorphic zones. Mosses and algae in the study area appear to be largely unknown, so to supplement information on biodiversity in the study area, a preliminary list of mosses and alga was compiled by Dr. R. Palmer as part of the aquatic macroinvertebrate survey (see Table 9 and Addendum A: Appendix B and C for more details).

Hillslope Seeps (BMU5: Valley bottom wetlands and seeps)

Hillslope Seepage Wetlands were common in the study area (Plate 1A). The seeps were characterised by peat soils and *Sphagnum* moss (Plate 1B). Aquatic biotopes in this zone comprised mostly shallow seeps that are likely to be active seasonally, and smaller areas that are likely to have permanent surface water. The filamentous Green alga *Stigeoclonium cf lubricum* was found in shallow seepage areas (Plate 1C). This genus is typically associated with fast-flowing, polluted streams, so its presence here was unexpected. The seepage areas also had patches of the floating protobacteria *Leptothrix discophora* (Plate 1D). This species is typically associated with oxidation of iron or manganese (Campbell 2003).

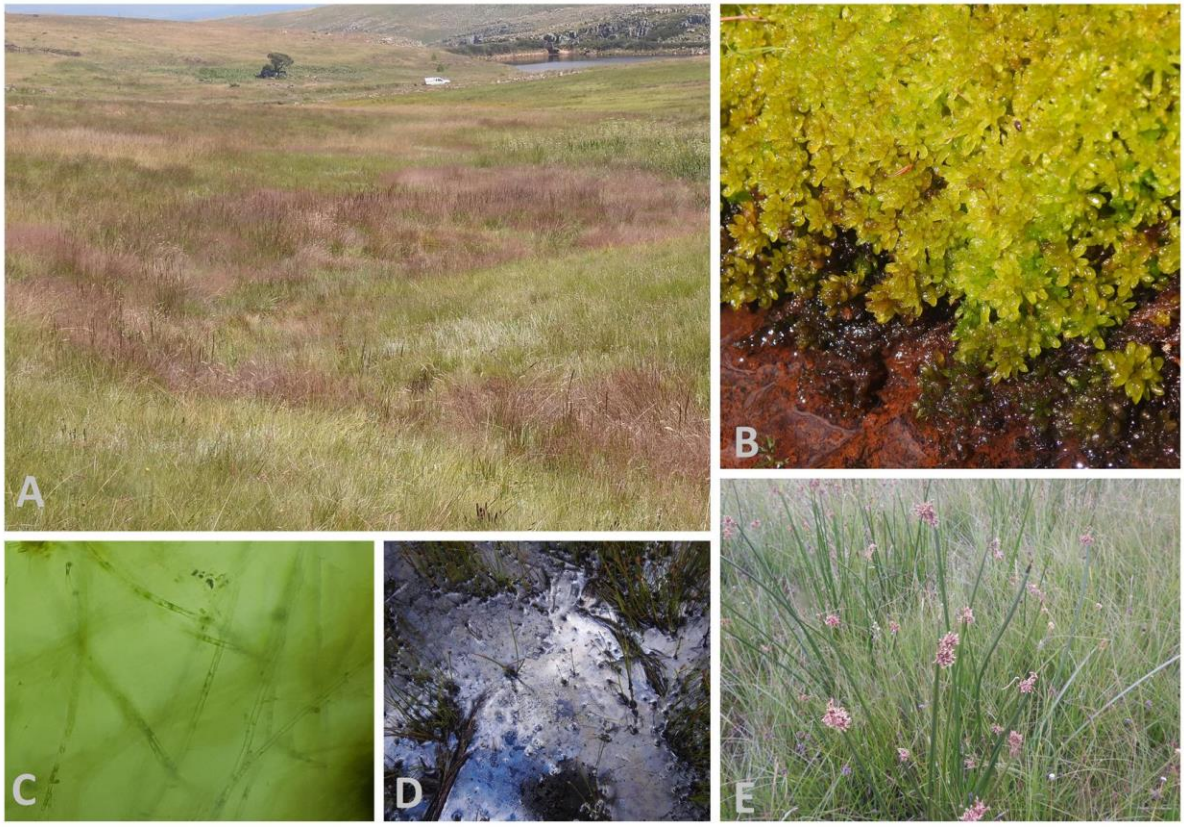

Plate 1: Hillslope Seep Zone

[A) Hillsope Seepage Wetland upslope of Site A1; B) *Sphagnum sp.* (Sphagnaceae); C) *Stigeoclonium cf. lubricum* (Chaetophoracea); D) *Leptothrix discophora* (Comamonadaceae); E) *Schoenoplectus brachyceras* (Cyperaceae).]

Source Zone (BMU5: Valley bottom wetlands and seeps)

Source Zones were common in the study area (Plate 2A). Aquatic biotopes in this zone comprised springs, bedrock sheets, pools and shallow runs. One of the most abundant aquatic plants in shallow runs within this zone was the Red alga *Batrachospermum sp.* (Plate 2B). This genus is typically associated with unpolluted bogs (Sheat and Vis, 2015). The shallow runs also contained high abundance of an unidentified bacterium (Plate 2C). A characteristic feature of the pools in this zone was the abundance of submerged *Isolepis fluitans* (Plate 2D). This species provided suitable substrate for aquatic macroinvertebrates. A spring that entered the Klip River tributary at Site C1 supported a dense growth of the filamentous alga *Mougeotia* sp. (Plate 2E). This genus is typically associated with acidic conditions associated with *Sphagnum* bogs (http://fmp.conncoll.edu/).

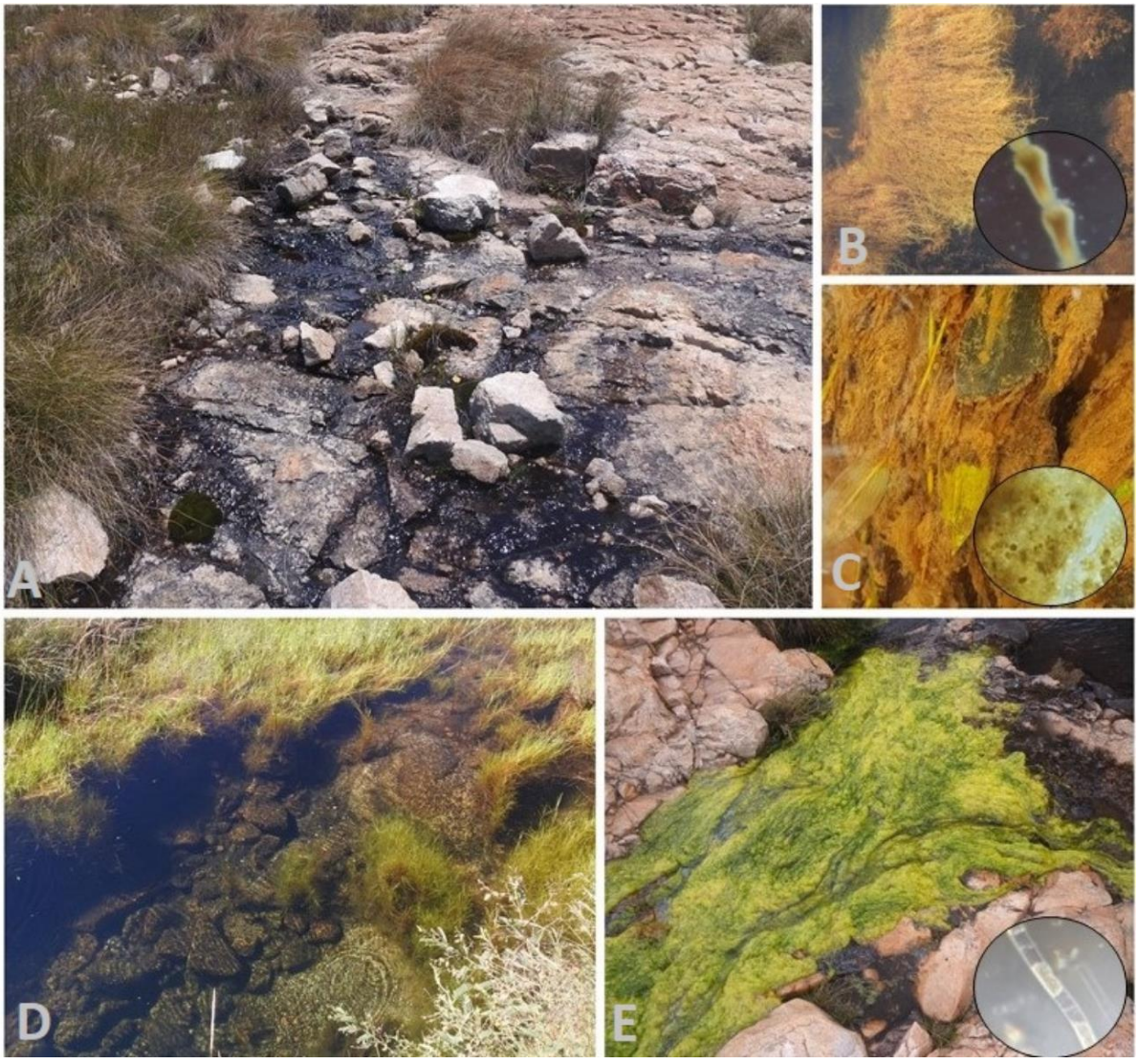

Plate 2: Source Zone

[A) Seep biotope within Source Zone; B) *Batrachospermum sp.* (Batrachospermaceae); C) Unidentified bacterium; D) *Isolepis fluitans* (Cyperaceae); E) *Mougeotia sp.* at Site C1 (Zygnemataceae).]

Mountain Headwaters (BMU7: Mountain streams)

Mountain Headwaters were common in the study area (Plate 3A). Aquatic biotopes in this zone comprised bedrock, pools, runs, riffles, cascades and waterfalls. In addition, splash zones of cascades and waterfalls created micro-habitat that were colonised by various species of moss and liverwort. Liverworts included *Symphogyna podophylla* (Plate 3B), *Fossombronia* sp. (Plate 3C), and *Marchantia* sp. (Plate 3D). Bedrock in-current also supported matts of the Red alga *Tuomeya* sp. (Plate 3D). This alga created small patches suitable for colonisation of various aquatic macroinvertebrates, such as baetid mayflies and Chironomidae.

Plate 3: Mountain Headwater Zone

[A) Cascade biotope within Mountain Headwater Zone; B) *Symphogyna podophylla* (Pallavicinaceae); C) *Fossombronia sp.* (Fossombroniaceae); D) *Marchantia sp.* (Marchantiaceae); E) *Tuomeya sp.* (Batrachospermaceae).]

Mountain streams (BMU7)

Mountain Streams were common in the Study Area (Plate 4A). Aquatic biotopes in this zone comprised bedrock, pools, runs, riffles and cascades. Stream bed composition was characterised by a wide range of particle sizes, including gravels, cobbles and boulders, and these provided ideal substrates for colonisation by aquatic macroinvertebrates. Mountain Streams in the Study Area supported little to no submerged or emergent vegetation. Marginal vegetation was characterised by shrubs such as *Cliffortia linearifolia* (Plate 4B), and *Morella microbracteata* (EN) (Plate 4C), and the geophyte *Hesperantha coccinea* (Plate 4D).

Plate 4: Mountain Stream Zone

[A&E) Run biotope within Mountain Stream Zone; B) *Cliffortia linearifolia* (Rosaceae); C) *Morella microbracteata (EN)* (Myriacaceae); D) *Hesperantha coccinea* (Iridaceae).]

Earth Dams (BMU 11: Dams)

There were two earth dams in the DBPNR within the Groot-Dwars River Catchment. Dam 1 covered ~1.0 hectares at full supply (Plate 5A). Dam 2 was smaller and covered ~0.4 hectares at full supply but was not surveyed for this report. Aquatic macrophytes at Dam 1 were dominated by the submerged *Lagarosiphon major* (Plate 5C), the floating *Potamogeton nodosus (*Plate 5D), and the presence of Bulrush *Typha capensis.* These plant species were not found in natural pools within the study area, so their presence is attributed to artificial habitat created by impoundment.

Seepage downstream of Dam 1 was characterised by dense growth of the protobacterium *Leptothrix ochracea* (Plate 5E). This bacterium clogged interstitial spaces and created conditions that were unsuitable for aquatic macroinvertebrates. This species is typically associated with oxidation of iron (Fleming *et al*. 2011). An unusual characteristic of such blooms is that over 90% of the sheaths are empty, so what appears as an abundant population comprises mostly moribund cells (Fleming *et al.* 2011).

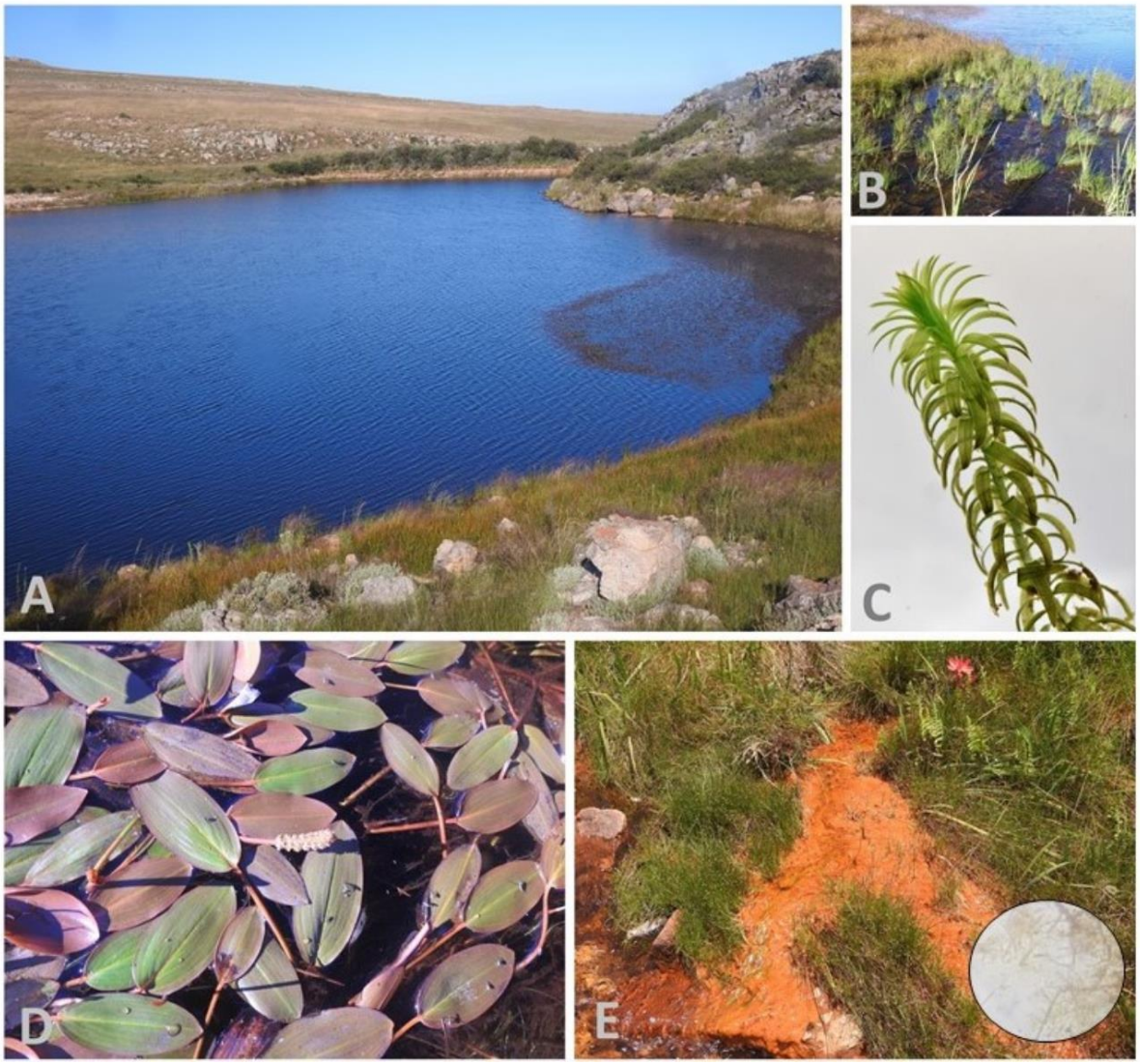

Plate 5: Earth Dams

[A) De Berg Dam 1; B) Inflow into de Berg Dam 1 near Site A2; C) *Lagarosiphon major* (Hydrocharitaceae); D) *Potamogeton nodosus* (Potamogetonaceae); E) *Leptothrix ochracea* (Comamondaceae).]

Table 9: List of aquatic plants recorded on DBPNR (Palmer, 2022).

Mosses (Bryophyta)	
Bartramiaceae	*Breutelia sp.*
Brachytheciaceae	*Rhynchostegium sp.*
Bryaceae	*Anomobryum julaceum*
Bryaceae	*Bryum cellulare*
Bryaceae	*Bryum pseudotriquetrum*
Bryaceae	*Philonotis sp.*
Ditrichaceae	*Ditrichum sp.*
Fissidentaceae	*Fissidens ovatus*
Leucobryaceae	*Campylopus sp.*
Polytrichaceae	*Polytrichum sp.*
Pottiaceae	*Barbula sp.*
Pottiaceae	*Weissia controversa*
Sphagnaceae	*Sphagnum sp.*
Liverworts (Marchantiophyta)	
Fossombroniaceae	*Fossombronia sp.*
Marchantiaceae	*Marchantia sp.*
Pallaviciniaceae	*Symphyogyna podophylla*
Ricciaceae	*Riccia sp.*
Red Algae (Rhodophyta)	
Batrachospermaceae	*Batrachospermum sp.*
Batrachospermaceae	*Nothocladus sp.*
Protobacteria	
Comamonadaceae	*Leptothrix discophora*
Comamonadaceae	*Leptothrix ochracea*
Green Algae (Chlorophyta)	
Zygnemataceae	*Mougeotia sp.*
Chaetophoraceae	*Stigeocloeon cf. lubricum*

4.4 Aquatic Macro-invertebrates

Aquatic macroinvertebrates are especially valuable indicators of water quality alteration in aquatic ecosystems. Macroinvertebrate sampling of stream sites are generally performed on family level using the South African Scoring System (SASS5) protocol but as part of the current study selected sites were earmarked for more detailed studies to initiate an inventory of aquatic macro-invertebrates on species level and to establish if any aquatic macroinvertebrate species of conservation concern are present within the DBPNR area. The SASS5 protocol (family levels) were applied at various sites between 2020 and 2022 (by Clean Stream Biological Services), while a once-off field survey was conducted by Dr. R. Palmer (Nepid) during February 2022 to collect primary data on aquatic macroinvertebrate species at representative sites in the streams and wetlands within the study area.

Aquatic macroinvertebrate species diversity and conservation status (Palmer, 2022: Addendum A)

Aquatic macroinvertebrates were collected using various methods, including baited traps and nets and high resolution photographs were taken in the field and used to identify taxa to species, where possible, or to the lowest taxonomic level feasible. The following groups were included by Palmer (2022):

Annelids	Crustaceans	Insects	Molluscs
- leaches	- crabs - clam shrimps - fairy shrimps - caridean shrimps	- beetles - bugs - caddisflies - dragonflies - damselflies - true flies - mayflies	- gastropods - bivalves

The field survey by Dr. R. Palmer was undertaken between 7 and 9 February 2022 and was conducted during wet "La Niña" weather conditions. Springs were active and stream levels were moderate and stable, so conditions were ideal for sampling aquatic macroinvertebrates. Aquatic ecosystems were classified according to hydrogeomorphic units described by Ollis *et al.* (2013). Aquatic biotopes and aquatic flora associated with each hydrogeomorphic zone were identified in the field. Aquatic macrophytes and mosses were photographed in the field using a Nikon Coolpix P900. Filamentous algae and bacteria were photographed in the field using a Galaxy Note 8 smart phone attached to a Newton Nm1 field microscope. Aquatic macroinvertebrates were collected from aquatic and aerial habitats as follows:

Aquatic

- physical removal and examination of submerged substrates (cobbles; marginal vegetation etc);
- SASS5 kick-net, swept through marginal and submerged vegetation; and
- small aquarium net, used for sampling shallow water, such as bedrock sheets.

Aerial

- butterfly net, used mainly along stream margins to collect adult true flies (Diptera), and damselflies and dragonflies (Odonata);
- light trap, comprising a portable LED work light (100W) suspended over a tray of water with drop of dish washing liquid. The trap was set overnight for one night only at a lodge some 3 km from the Study Area. The trap was intended to collect flying aquatic insects that are attracted to lights, such as caddisflies and mayflies;
- malaise trap baited with octenol (ox breathe), set for ~70 hours at Site A1. The trap was intended to collect blood-sucking aquatic insects, such as horseflies and mosquitoes.

The reason for collecting adult stages of flying aquatic insects was because many species are described as adult males only. Most specimens were identified in the field. Identifications were based on naked eye examination, or with a 10x loupe. Specimens that could not be identified in the field were preserved in 70% ethanol for more detailed examination under a dissecting microscope. Representative specimens were euthanised with ethyl acetate (adults), 70% ethanol (larvae), or soda water (annelida), and then photographed in the field using a Nikon D750 with a 200 mm macro lens.

Hillslope Seepage Wetlands (BMU5)

Hillslope Seepage Wetlands supported a low diversity of macroinvertebrates, with eight taxa recorded. Sampling intensity in these wetlands was low because of the limited availability of aquatic habitats, and because no attempt was made to sample the underlying peat soils. The diversity of aquatic macroinvertebrates in these wetlands is likely to be significantly higher than reported here. The most notable feature of these wetlands was the high abundance of *Tipula* species (Tipulidae), and the presence of Psychodid flies (Psychodidae), and the Marble-eye hoverfly *Eristalinus modestus* (Syrphidae), all of which reflect the high organic content of the wetland soils.

Source Zone (BMU5)

The Source Zone supported a high diversity of macroinvertebrates, with 37 taxa recorded. Sampling in this zone was focused on the two tributaries that fed Dam 1 (i.e. Sites A1 and A2). The diversity of aquatic macroinvertebrates in this zone is likely to be higher than reported here. Ten taxa that are sensitive to water quality deterioration were recorded in this zone. Sensitive taxa included the stonefly *Afronemoura stuckenbergi*, which is endemic to mountain streams in Mpumalanga Province, and the rare beetle *Torrendincola rhodesica* (Torrindincolidae), which is endemic to mountain streams in Mpumalanga Province and Zimbabwe (Villet and Endrödy-Younga 2007). The zone also supported an unusually high abundance of the leach *Hemiclepsis* sp. (Glossiphonidae), and the flatworm *Dugesia* sp. (Dugesiidae). Another notable taxon recorded in the zone was the blackfly *Simulium bequaerti,* which is typically associated with small, clear temporary streams (Palmer and de Moor, 1991).

Mountain Headwaters (BMU7)

Mountain Headwaters supported a high diversity of macroinvertebrates, with 43 taxa recorded. Sampling in this zone was focused on cascades and waterfalls on the Groot-Dwars River (Site A4) and Everest Tributary (Site B1). The comparatively high diversity of macroinvertebrates recorded in this zone is attributed partly to

the presence of waterfalls, as some species, such as *Simulium debegene* (Simuliidae) are restricted to waterfalls, and partly because the sampling intensity in this zone was higher than the other zones. Eleven taxa that are sensitive to water quality deterioration were recorded. Sensitive taxa included the Net-wing midge *Elporia marieps* (Blephariceridae), which is endemic to mountain streams in Mpumalanga Province (Stuckenberg 1961). The presence of the baetid mayfly *Demoreptus cf monticola*, indicated that biological water quality was good (Barber-James and Lugo-Ortiz, 2003). The genus *Demoreptus* comprises three known species that are restricted to mountain streams in southern Africa. However, a genetic study showed that the genus contains additional undescribed cryptic species associated with range-restricted distribution (Taylor, 2015).

Mountain Streams (BMU7)

Mountain Streams supported a lower diversity of macroinvertebrates (34 taxa) than Mountain Headwaters (43 taxa). The lower diversity is attributed mainly to the absence of waterfalls in this zone. A notable feature of this zone was high abundance of the adult Mountain malachite *Chlorolestes fasciatus.* This species is restricted to high lying escarpment areas of Mpumalanga, KwaZulu-Natal and Eastern Cape Provinces (Tarboton and Tarboton 2005). Apart from differences attributed to waterfalls, the composition of taxa in Mountain Streams and Mountain Headwater was much the same, and not discussed further.

Earth Dams (BMU11)

Dam 1 supported a moderate diversity of macroinvertebrates, with 17 taxa recorded in around the dam. Sampling intensity at the dam was low because of the limited likelihood of finding species of conservation interest, and because no attempt was made to survey planktonic micro-crustaceans that are likely to occur in the impounded water. The dam supported taxa that are typically associated with standing water, such as Chironomidae, Ceratopogonidae, Gerridae, Naucoridae and Pleidae. High numbers of adult Swamp bluet *Africallagma glaucum* (Coenagrionidae) were recorded around Dam 1.

Table 10: List of Aquatic Macroinvertebrates sampled at selected sites in DBPNR (Palmer, 2022).

Taxon (sensitivity)	Taxon	Status	Seep	Source	M. Headwater	M. Stream	Earth Dam
ANNELIDA							
Glossiphoniidae	*Hemiclepsis sp.*	-	-	B	A	-	-
Glossiphoniidae	*Unidentified sp.*	-	-	1	-	-	-
PLATYHELMENTHES							
Dugesiidae	*Dugesia sp.*	-	A	B	A	-	-
ARACHNIDA							
Hydrachnidae	*Hydrachna sp.*	-	-	-	-	-	1
CRUSTACEA							
Potamonautidae (3)	*Potamonautes sidneyi*	LC	-	-	A	A	-
ENTOGNATHA							
Collembolla	-	-	A	-	-	-	-
PLECOPTERA							
Notonemouridae (14)	*Afronemoura stuckenbergi*	-	-	B	B	B	-
EPHEMEROPTERA							
Baetidae	*Species A*	-	-	-	1	1	-
Baetidae	*Baetis harrisoni*	-	-	A	A	A	-
Baetidae	*Demoreptus cf monticola*	-	-	A	B	B	-
Leptophlebiidae (9)	*Adenophlebia sp.*	-	-	A	A	A	-
Tricorythidae (9)	*Trichorythus discolor*	-	-	-	1	A	-
ANISOPTERA							
Aeshnidae (8)	*Anax sp.*	LC	-	1	A	A	-
Gomphidae (6)	*Notogomphus praetorius*	LC	-	-	-	1	-
Libellulidae (4)	*Crocothemis sanguinolenta*	LC	-	-	-	1	-
Libellulidae (4)	*Orthetrum caffrum*	LC	1	A	B	-	-
Libellulidae (4)	*Sympetrum fonscolombii*	LC	-	-	-	-	1
Libellulidae (4)	*Trithemis stictica*	LC	-	A	B	A	A
ZYGOPTERA							
Coenagrionidae (4)	*Africallagma glaucum*	LC	-	B	B	B	B
Coenagrionidae (4)	*Pseudagrion caffrum*	LC	-	A	B	-	-
Coenagrionidae (4)	*Pseudagrion spernatum*	LC	-	1	-	-	-
Platycnemididae (10)	*Allocnemis leucosticta*	LC	-	-	B	B	-
Platycnemididae (10)	*Elattoneura glauca*	LC	-	A	A	-	-
Synlestidae (8)	*Chlorolestes fasciatus*	LC	-	B	B	A	1
HEMIPTERA							
Corixidae (3)	-	-	-	-	-	-	A
Gerridae (5)	SF: Gerinae	-	-	A	A	A	A
Naucoridae (7)	*Species A*	-	-	A	A	A	A
Naucoridae (7)	*Laccocoris sp.*	-	-	A	A	A	A
Notonectidae (3)	*Enithares sp.*	-	-	-	-	-	A
Pleidae (4)	*Paraplea sp.*	-	-	-	-	-	A
TRICHOPTERA							
Hydropsychidae (4)	*Cheumatopsyche sp.*	-	-	A	A	A	-
Hydroptilidae (6)	*Hydroptila crutiata*	-	-	A	A	A	-
Lepidostomatidae (10)	*Goerodes sp.*		-	A	A	A	-
Leptoceridae (6)	*Athripsodes harrisoni*	-	-	A	A	A	-
Leptoceridae (6)	*Setodes sp.*	-	-	B	A	A	-

Table 10: Continued

Taxon (sensitivity)	Taxon	Status	Seep	Source	M. Headwater	M. Stream	Earth Dam
COLEOPTERA							
Dytiscidae (5)	*Laccophilus inornatus*	-	-	A	-	-	-
Dytiscidae (5)	*Laccophilus lineatus*	-	-	1	-	-	-
Dytiscidae (5)	*Neptosternus sp.*	-	-	1	-	-	-
Dytiscidae (5)	*Rhantaticus congestus*	-	-	-	-	-	1
Elmidae (8)	*Pseudancyronyx sp.*	-	-	A	A	A	-
Gyrinidae (5)	*Aulonogyrus sp.*	-	-	A	A	A	-
Gyrinidae (5)	*Dineutus sp.*	-	-	A	A	A	-
Scirtidae (12)	-	-	-	1	-	-	-
Torrindincolidae	*Torridincola rhodesica*	-	-	1	-	-	-
NEMATOCERA							
Blephariceridae (15)	*Elporia marieps*	-	-	-	B	A	-
Ceratopogonidae (5)	*Bezzia sp.*	-	-	-	-	-	1
Chironomidae (2)	-	-	A	A	A	A	A
Culicidae (1)	*Culex sp.*	-	-	-	-	-	1
Dixidae (10)		-	-	A	-	-	-
Psychodidae (1)	-	-	A	-	-	-	-
Simuliidae (5)	*Simulium bequaerti*	-	-	A	A	A	-
Simuliidae (5)	*Simulium debegene*	-	-	-	B	A	-
Simuliidae (5)	*Simulium dentulosum*	-	-	-	B	B	-
Simuliidae (5)	*Simulium meduaseforme*	-	-	-	A	B	-
Simuliidae (5)	*Simulium nigritarse*	-	-	A	A	A	-
Tipulidae (5)	*Erioptera sp.*	-	A	-	-	-	-
Tipulidae (5)	*Tipula sp.*	-	C	-	-	-	-
BRACHYCERA							
Anthomyiidae	*Anthomyia (punctipennis) sp.*	-	-	-	1	-	-
Anthomyiidae	*Anthomyia concava*	-	-	A	A	A	1
Dolichopodidae	*Medetera sp.*	-	-	-	A	A	-
Empididae (6_	*Empis sp.*	-	-	-	1	-	-
Ephydridae (3)	*Paralimna sp.*	-	-	-	A	-	-
Rhagionidae	*Atherimorpha sp.*	-	-	-	1	-	-
Syrphidae (1)	*Eristalinus modestus*	-	A	A	-	-	-
Tachinidae	-	-	-	-	A	-	-

Aquatic macroinvertebrates families: South African Scoring System (SASS5)

The aquatic macro-invertebrate community of the study area was also assessed on a family level at selected sites by applying the SASS5 (South African Scoring System, version 5) protocol (Tables 11 to 13). These baseline SASS5 and ASPT will also be valuable to measure any future changes to the aquatic ecosystems within the DBPNR. A total of **47 invertebrate families were recorded** from all sampling sites in the DBPNR study area from 2020 to 2022 (Tables 11 and 12). This diversity (number of taxa) was relatively high for these upper reaches, source zones and wetland systems. The presence of five taxa with a high requirement for good water quality (Blepharoceridae, Notonemouridae, Perlidae, >2 species of Baetidae and Pyralidae, together with another eleven taxa with a moderate requirement for unmodified water quality confirmed that the general water quality of the DBPNR is currently very good to pristine. A high proportion of the invertebrate taxa are also habitat specialists. These are at risk to any activities which result in the deposition of eroded sediments within the stream channel (such as over grazing, construction).

The total SASS5 scores ranged between 185 (Groot Dwars River at site GD-SterkfonteinP3) to 59 (Potspruit at site PS1). The ASPT values ranged between 7.6 (Everest tributary at site Everest 3) to 4.9 (Potspruit at site PS1 and Groot Dwars River at site GD_De Berg). The high SASS5 and ASPT values furthermore indicate good water quality and overall biotic conditions prevailing in some streams of the study area. The

variation in SASS5 scores and ASPT values furthermore confirm variation in the invertebrate composition between different sites and streams which are attributed to natural variation (habitat differences) as well as potential response to human impacts (such as flow modification, erosion, water quality deterioration).

Groot Dwars River

A SASS5 score of 97 and ASPT of 5.4 was recorded at site US-Dam 1 (BMU5: Valley bottom wetlands and seeps) in the most upper Groot Dwars River directly upstream of GD Dam1 in close proximity to its source (Table 7). The SASS5 score (93) and ASPT (4.9) decreased slightly downstream towards site GD_De Berg (BMU7: Mountain Stream) directly downstream of De Berg Dam1) (Table 12). The river downstream of De Berg Dam 1 supported high abundance of the blackfly *Simulium meduaseforme* (Simuliidae), a species typically associated with impoundment outlets, and its presence in high abundance downstream of Dam 1 reflects an alteration in the trophic structure of the river caused by plankton discharged from the impounded (Nepid, 2022). Seepage downstream of the dam was also characterised by dense growth of the protobacterium *Leptothrix ochracea* (Nepid, 2022). This bacterium clogged interstitial spaces and created conditions that were unsuitable for aquatic macroinvertebrates. This species is typically associated with oxidation of iron (Fleming *et al.* 2011). An unusual characteristic of such blooms is that over 90% of the sheaths are empty, so what appears as an abundant population comprises mostly moribund cells (Fleming *et al.* 2011). The notable impacts in this area include the dam and old farmstead, now converted to offices, and road (leading to communication tower). The SASS5 score (185) and ASPT (6.4) increased notably downstream towards site GD_SterkfonteinP3, indicating excellent conditions prevailing in this Groot Dwars River reach within DBPNR (Table 12). The high ASPT especially reflects very good water quality, further confirmed by the presence of two families with a very high requirement for unmodified water quality (Perlidae and Blepharoceridae). The SASS5 score of 93 measured in the Northern tributary of the Groot Dwars River (site GD N-trib: BMU7: Mountain stream) compared well with the other upstream sites, while the high ASPT (6.6) confirmed good water quality prevailing. Habitat modifiers observed in the Northern tributary catchment included the presence of alien vegetation (Black wattle), bank erosion and limited grazing by livestock.

Everest Tributary

A SASS5 score of 89 and ASPT of 6.4 was recoded at site Everest 1 (BMU5: Wetlands and seeps) in the most upper reaches of Everest Tributary, reflecting very good water quality (based on ASPT) (Table 13). Condition improved downstream towards sites Everest 2 (SASS5: 141 and ASPT: 5.9) and Everest 3 (SASS5: 136 and ASPT: 7.6) (both in BMU7: Mountain streams) confirming very good biotic conditions and especially good water quality prevailing throughout the Everest Tributary within the DBPNR. The conditions in the upper reaches (inside De Berg) of the Everest Tributary is thought to be excellent (near pristine) with only minor impacts in this catchment (alien vegetation stand, Bluegum), erosion (roads/fire breaks).

Klip River catchment

In the unnamed tributary of the Klip River falling inside DBPNR (sites KR1 and KR2: BMU7: Mountain streams) the SASS5 scores ranged between 83 and 98, with ASPT values of 6.4 and 6.1 measured (Table 14). Since no notable impacts occur within this upper catchment (apart from road and satellite towers) these values can be estimated to reflect near-natural conditions. The presence of two taxa with a high requirement for unmodified water quality (Blepharoceridae and Notonemouridae) and a further four taxa with a moderate requirement for unmodified water quality confirms that good water quality is currently prevailing in this stream. A spring that entered the Klip River tributary at Site KR1 supported a dense growth of the filamentous alga *Mougeotia* sp., a genus typically associated with acidic conditions associated with *Sphagnum* bogs (http://fmp.conncoll.edu/) (Nepid, 2022).

Potspruit

A relatively low SASS5 score (59) and ASPT (4.9) was calculated for site PS1 (BMU5: Wetlands and seeps) in the Potspruit falling within the Goedehoop farm section of the DBPNR (Table 13). This site is below farm dams and a high level of erosion caused significant sedimentation that impacted notably on the bottom substrates (clogging interstitial spaces) and hence affecting the aquatic invertebrates assemblage negatively. The high level of erosion may be associated with altered veld condition (over grazing, trampling, altered fire regime).

Table 11: SASS5 analysis, including macro-invertebrate families sampled, ASPT, water quality, flow and cover preferences of the Groot Dwars River sites within the DBPNR study area (2020-2022).

Taxon	Common name	US-Dam1				GD_De Berg				GD_SterkfonteinP3				GD N-trib				Flow (in m/s) preference				Cover preference				
		Stones	Veg	GSM	Total	Stones	Veg	GSM	Total	Stones	Veg	GSM	Total	Stones	Veg	GSM	Total	<0.1	0.1-0.3	0.3-0.6	>0.6	BEDROCK	COBBLES	VEG	GSM	WATER COLUMN
Blepharoceridae	Net-winged midges	-	-	-	-	-	-	-	-	A	-	-	A	B	-	-	B	0	0	3	4	2	3	0	0	0
Notonemouridae	Stoneflies	A	A	A	A	-	-	-	-	-	-	-	-	-	-	-	-	1	1	2	4	1	4	1	0	0
Perlidae	Stoneflies	-	-	-	-	-	-	-	-	A	A	A	B	-	-	-	-	1	1	1	5	1	4	1	0	0
Baetidae > 2 spp.	Small minnow flies	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	B									
Platycnemidae	Damselflies	-	-	-	-	-	-	-	-	-	-	-	-	-	1	A	A	2	3	1	1	0	2	3	0	0
Philopotamidae	Caseless caddisflies	-	-	-	-	-	-	-	-	A	-	-	A	-	-	-	-	0	1	2	3	1	4	1	1	0
Psephenidae	Water penny beetles	-	-	-	-	-	-	-	-	1	-	-	1	-	-	-	-	0	1	3	4	1	4	1	0	0
Athericidae	Snipe flies	-	-	-	-	-	-	-	-	A	A	-	A	-	-	-	-	0	1	2	2	1	4	1	1	0
Leptophlebiidae	Prongills	A	-	A	B	-	-	-	-	1	1	B	B	A	A	A	B	3	2	2	1	1	3	2	0	0
Tricorythidae	Stout crawlers	-	-	-	-	-	-	-	-	A	-	-	A	-	-	-	-	0	1	1	4	1	4	1	0	0
Chlorolestidae	Damselflies	-	1	A	A	-	-	-	-	-	-	-	-	-	-	-	-	3	2	1	0	0	1	4	0	0
Lestidae	Damselflies	-	-	-	-	-	-	1	1	-	-	1	1	-	-	-	-	4	1	0	0	0	1	4	1	0
Aeshnidae	Dragonflies	A	A	1	A	-	-	-	-	A	-	-	A	A	-	-	A	1	2	2	2	0	3	2	0	0
Ecnomidae	Caseless caddisflies	-	-	-	-	-	-	1	1	-	-	-	-	-	-	-	-	1	5	0	0	2	3	2	0	0
Naucoridae*	Creeping water bugs	-	-	-	-	A	A	-	A	A	A	-	A	-	-	-	-	2	2	3	0	1	1	1	1	4
Baetidae 2 spp.	Small minnow flies	B	-	-	B	A	-	-	A	B	-	B	B	B	B	A	B									
Caenidae	Cainflies	-	-	-	-	-	-	1	1	A	A	-	A	-	-	-	-	3	2	1	1	0	2	1	3	0
Gomphidae	Dragonflies	-	-	-	-	-	A	A	A	A	-	A	B	-	-	-	-	0	2	3	0	0	1	0	5	0
Hydropsychidae 2 spp.	Caseless caddisflies	-	-	-	-	-	-	-	-	B	-	-	B	-	-	-	-									
Hydroptilidae	Micro caddisflies	-	-	-	-	-	-	-	-	-	A	B	B	-	-	-	-	0	3	2	2	1	2	3	1	0
Leptoceridae	Cased caddisflies	A	A	1	B	1	B	A	B	-	A	-	A	-	A	-	A	0	1	3	2	2	2	2	2	0
Ancylidae	Limpets	-	-	-	-	-	-	-	-	A	-	-	A	-	-	-	-	1	2	2	1	3	2	1	0	0
Gerridae*	Pond skater	-	A	-	A	-	-	-	-	-	-	-	-	-	-	-	-	4	1	0	0	0	0	0	0	5
Veliidae*	Broad-shouldered water strid	A	A	-	A	-	A	-	A	-	-	1	1	-	-	-	-	5	1	1	0	0	0	0	0	5
Dytiscidae (adults*)	Predacious diving beetles	A	-	A	A	-	A	-	A	-	-	A	A	-	A	-	A	4	2	1	0	1	2	3	1	2
Gyrinidae (adults*)	Whirligig beetles	-	-	A	A	A	B	A	B	B	A	A	B	A	A	A	B	1	2	2	3	0	0	0	0	5
Ceratopogonidae	Biting midges	A	A	A	B	-	-	-	-	A	A	A	B	-	-	A	A	2	2	2	4	2	3	2	2	0
Simuliidae	Black flies	-	A	1	A	B	1	-	B	B	A	-	B	B	-	-	B	0	2	2	4	2	3	2	0	0
Tabanidae	Horseflies	-	-	-	-	-	-	1	1	1	-	1	A	-	-	-	-	2	3	1	0	0	2	0	3	0
Tipulidae	Crane flies	-	-	-	-	1	-	-	1	-	-	-	-	-	-	-	-	3	4	1	1	1	2	0	3	0
Baetidae 1 sp.	Small minnow flies	-	A	-	A	-	-	-	-	-	A	-	A	-	-	-	-	2	2	2	2	2	2	2	2	1
Coenagrionidae	Damselflies	-	A	1	A	-	-	-	-	-	A	-	A	1	B	A	B	1	2	3	1	0	1	4	1	0
Libelludae	Dragonflies	-	-	-	-	-	A	A	A	-	-	-	-	-	-	-	-	1	2	3	1	1	4	0	1	0
Hydropsychidae 1sp.	Caseless caddisflies	-	-	-	-	-	-	-	-	-	-	-	-	A	-	-	A	0	1	2	4	2	3	1	0	0
TURBELLARIA	Flatworms	B	-	-	B	-	-	-	-	-	-	-	-	-	-	-	-	1	2	3	4	1	4	0	0	0
Leeches	Leaches	-	-	-	-	-	1	-	1	-	-	-	-	-	-	-	-	2	2	1	1	0	4	1	1	0
Potamonautidae*	Crabs	-	-	-	-	-	-	-	-	1	-	-	1	1	-	-	1	1	1	3	2	0	3	1	1	0
Corixidae*	Water boatmen	-	-	-	-	A	-	-	A	-	-	A	A	-	-	-	-	2	3	1	0	1	1	1	1	4
Nepidae*	Water scorpions	-	1	-	1	-	-	-	-	-	-	-	-	-	-	-	-	4	1	0	0	0	0	5	0	0
Notonectidae*	Back swimmers	-	A	-	A	-	A	-	A	-	A	-	A	-	-	-	-	4	1	0	0	0	0	2	0	4
Chironomidae	Midges	A	A	A	B	A	A	B	B	-	-	-	-	A	A	A	A	1	3	2	2	2	2	2	2	0
Oligochaeta	Aquatic earthworms	-	-	-	-	-	-	1	1	A	-	-	A	-	-	-	-	2	2	2	1	0	1	0	4	0
Culicidae*	Mosquitoes	A	A	1	A	-	-	-	-	1	-	-	1	-	-	-	-	3	1	0	0	0	0	0	0	5
Total SASS5 score		64	73	72	97	39	51	51	93	145	82	75	185	61	47	41	93									
No. of families		11	14	12	18	8	11	10	19	21	13	12	29	10	8	7	14									
ASPT		5.82	5.21	6.00	5.39	4.88	4.64	5.10	4.89	6.90	6.31	6.25	6.38	6.10	5.88	5.86	6.64									
Total IHAS					80				91				98				90									
IHAS - Habs sampled					39				47				54				49									
IHAS - Stream condition					41				44				44				41									
Suitability score		4	10	9	23	6	3	9	18	12	10	7	29	8	2	6	16									

Key: High requirement for unmodified water quality; Veg=Vegetation

Key:	Preference
	0 - No preference (does not o
	1 - Very low preference Coinc
	2 - Low preference
	3 - Moderate preference
	4 - High preference
	5 - Very high preference

Key: High requirement for unmodified water quality; Veg=Vegetation

Moderate requirement for unmodified water quality

Low requirement for unmodified water quality

Very low requirement for unmodified water quality

A = 1-10 individuals; B = 11-100 individuals; C = 101-1000 individuals; ASPT = Average score per taxon.

Table 12: SASS5 analysis, including macro-invertebrate families sampled, ASPT, water quality, flow and cover preferences of the Everest Tributary sites within the DBPNR study area (2020-2022).

Taxon	Common name	Everest 1				Everest 2				Everest 3			
		Stones	Veg	GSM	Total	Stones	Veg	GSM	Total	Stones	Veg	GSM	Total
Blepharoceridae	Net-winged midges	-	-	-	-	1	-	-	1	B	-	-	B
Notonemouridae	Stoneflies	-	-	-	-	-	A	-	A	B	A	-	B
Baetidae > 2 spp.	Small minnow flies	B	-	-	B	B	B	-	B	-	-	-	B
Pyralidae	Aquatic caterpillars	-	-	-	-	-	-	-	-	A	-	-	A
Philopotamidae	Caseless caddisflies	1	-	-	1	1	-	-	1	1	-	-	1
Leptophlebiidae	Prongills	A	-	-	A	B	A	1	B	B	1	-	B
Tricorythidae	Stout crawlers	A	-	-	A	-	-	-	-	A	-	-	A
HYDRACARINA	Water mites	A	-	-	A	-	-	-	-	-	-	-	-
Chlorolestidae	Damselflies	-	-	-	-	-	-	-	-	A	A	A	A
Lestidae	Damselflies	A	A	-	A	-	-	-	-	-	-	-	-
Aeshnidae	Dragonflies	1	-	-	1	-	-	-	-	-	-	-	-
Naucoridae*	Creeping water bugs	-	-	-	-	-	A	1	A	A	A	A	B
Baetidae 2 spp.	Small minnow flies	-	-	-	-	-	-	A	A	B	-	-	B
Caenidae	Cainflies	-	-	-	-	A	-	A	B	-	-	-	-
Leptoceridae	Cased caddisflies	-	-	-	-	-	B	B	B	-	A	-	A
Ancylidae	Limpets	-	-	-	-	-	-	-	-	-	1	-	1
Veliidae*	Broad-shouldered water strid	-	-	-	-	-	1	-	1	-	-	-	-
Dytiscidae (adults*)	Predacious diving beetles	A	A	-	B	A	B	B	B	-	A	A	B
Gyrinidae (adults*)	Whirligig beetles	A	A	-	B	A	A	-	A	-	A	A	B
Hydrophilidae (adults*)	Water scavenger beetles	-	-	-	-	-	1	-	1	-	-	-	-
Ceratopogonidae	Biting midges	-	-	-	-	A	-	-	A	-	-	-	-
Simuliidae	Black flies	A	-	-	A	A	A	-	B	B	-	-	B
Tabanidae	Horseflies	-	-	-	-	-	-	-	-	1	-	-	1
Tipulidae	Crane flies	-	-	-	-	1	-	-	1	-	-	-	-
Baetidae 1 sp.	Small minnow flies	-	-	-	-	-	-	-	-	-	A	-	A
Coenagrionidae	Damselflies	-	A	-	A	A	B	A	B	-	-	-	-
Libelludae	Dragonflies	-	-	-	-	-	1	-	1	-	-	-	-
Hydropsychidae 1sp.	Caseless caddisflies	-	-	-	-	A	-	-	A	-	-	-	-
TURBELLARIA	Flatworms	-	-	-	-	A	1	A	A	-	-	-	-
Potamonautidae*	Crabs	-	-	-	-	-	-	1	1	1	-	-	1
Nepidae*	Water scorpions	-	-	-	-	-	1	-	1	-	-	-	-
Notonectidae*	Back swimmers	1	B	A	B	-	A	A	B	-	A	-	A
Chironomidae	Midges	A	A	-	A	A	A	A	B	A	-	A	A
Oligochaeta	Aquatic earthworms	-	-	-	-	-	-	1	1	-	-	-	-
Muscidae	House flies	-	-	A	A	-	-	-	-	-	-	-	-
Total SASS5 score		84	27	4	89	90	92	55	141	105	67	27	136
No. of families		12	6	2	14	14	16	12	24	13	10	5	18
ASPT		7.00	4.50	2.00	6.36	6.43	5.75	4.58	5.88	8.08	6.70	5.40	7.56
Total IHAS					87				85				96
IHAS - Habs sampled					45				46				54
IHAS - Stream condition					42				39				42
Suitability score		8	4	5	17	8	2	5	15	7	4	6	17

Flow (in m/s) preference				Cover preference				
<0.1	0.1-0.3	0.3-0.6	>0.6	BEDROCK	COBBLES	VEG	GSM	WATER COLUMN
0	0	3	4	2	3	0	0	0
1	1	2	4	1	4	1	0	0
1	1	3	2	0	2	3	0	0
0	1	2	3	1	4	1	1	0
3	2	2	1	1	3	2	0	0
0	1	1	4	1	4	1	0	0
0	2	2	0	1	1	2	3	1
3	2	1	0	0	1	4	0	0
4	1	0	0	0	1	4	1	0
1	2	2	2	0	3	2	0	0
2	2	3	0	1	1	1	1	4
3	2	1	1	0	2	1	3	0
0	1	3	2	2	2	2	2	0
1	2	2	1	3	2	1	0	0
5	1	1	0	0	0	0	0	5
4	2	1	0	1	2	3	1	2
1	2	2	3	0	0	0	0	5
0	2	2	0	0	0	3	2	2
2	2	2	4	2	3	2	2	0
0	2	2	4	2	3	2	0	0
2	3	1	0	0	2	0	3	0
3	4	1	1	1	2	0	3	0
2	2	2	2	2	2	2	2	1
1	2	3	1	0	1	4	1	0
1	2	3	1	1	4	0	1	0
0	1	2	4	2	3	1	0	0
1	2	3	4	1	4	0	0	0
1	1	3	2	0	3	1	1	0
4	1	0	0	0	0	5	0	0
4	1	0	0	0	0	2	0	4
1	3	2	2	2	2	2	2	0
2	2	2	1	0	1	0	4	0
4	2	2	0	1	1	1	1	4

Key: **Preference**
0 - No preference (does not o
1 - Very low preference Coinc
2 - Low preference
3 - Moderate preference
4 - High preference
5 - Very high preference

Key:

High requirement for unmodified water quality	Veg=Vegetation
Moderate requirement for unmodified water quality	
Low requirement for unmodified water quality	
Very low requirement for unmodified water quality	

A = 1-10 individuals; B = 11-100 individuals; C = 101-1000 individuals; ASPT = Average score per taxon.

Table 13: SASS5 analysis, including macro-invertebrate families sampled, ASPT, water quality, flow and cover preferences of the Klip River tributary and Potspruit sites within the DBPNR study area (2022).

Taxon	Common name	KR1				KR2				PS1	Flow (in m/s) preference				Cover preference				
		Stones	Veg	GSM	Total	Stones	Veg	GSM	Total	Total	<0.1	0.1-0.3	0.3-0.6	>0.6	BEDROCK	COBBLES	VEG	GSM	WATER COLUMN
Blepharoceridae	Net-winged midges	B	1	-	B	-	-	-	-	-	0	0	3	4	2	3	0	0	0
Notonemouridae	Stoneflies	B	-	-	B	B	B	-	B	-	1	1	2	4	1	4	1	0	0
Leptophlebiidae	Prongills	-	-	-	-	A	A	-	B	1	3	2	2	1	1	3	2	0	0
Tricorythidae	Stout crawlers	-	-	-	-	A	-	-	A	-	0	1	1	4	1	4	1	0	0
HYDRACARINA	Water mites	-	-	-	-	-	-	-	-	A	0	2	2	0	1	1	2	3	1
Chlorolestidae	Damselflies	A	-	A	A	A	A	A	B	-	3	2	1	0	0	1	4	0	0
Aeshnidae	Dragonflies	-	-	-	-	A	A	-	A	-	1	2	2	2	0	3	2	0	0
Naucoridae*	Creeping water bugs	A	A	-	A	-	A	-	A	-	2	2	3	0	1	1	1	1	4
Baetidae 2 spp.	Small minnow flies	-	-	-	-	-	-	-	A	-									
Leptoceridae	Cased caddisflies	A	-	-	A	-	-	-	-	A	0	1	3	2	2	2	2	2	0
Veliidae*	Broad-shouldered water strid	-	-	-	-	-	-	-	-	A	5	1	1	0	0	0	0	0	5
Dytiscidae (adults*)	Predacious diving beetles	A	A	A	B	A	A	A	B	A	4	2	1	0	1	2	3	1	2
Gyrinidae (adults*)	Whirligig beetles	A	A	-	B	A	A	-	B	A	1	2	2	3	0	0	0	0	5
Ceratopogonidae	Biting midges	-	-	A	A	-	-	1	1	-	2	2	2	4	2	3	2	2	0
Simuliidae	Black flies	A	A	-	A	A	A	-	A	A	0	2	2	4	2	3	2	0	0
Tipulidae	Crane flies	-	-	-	-	1	-	-	1	-	3	4	1	1	1	2	0	3	0
Baetidae 1 sp.	Small minnow flies	A	-	A	A	A	-	A	A	A	2	2	2	2	2	2	2	2	1
Coenagrionidae	Damselflies	-	A	-	A	-	A	-	A	-	1	2	3	1	0	1	4	1	0
Libelludae	Dragonflies	-	-	-	-	-	-	-	-	B	1	2	3	1	1	4	0	1	0
TURBELLARIA	Flatworms	-	-	-	-	-	-	-	-	A	1	2	3	4	1	4	0	0	0
Potamonautidae*	Crabs	-	-	-	-	1	-	-	1	-	1	1	3	2	0	3	1	1	0
Corixidae*	Water boatmen	-	-	-	-	-	-	-	-	A	2	3	1	0	1	1	1	1	4
Notonectidae*	Back swimmers	-	A	-	A	A	A	A	B	-	4	1	0	0	0	0	2	0	4
Chironomidae	Midges	-	-	A	A	A	1	1	A	A	1	3	2	2	2	2	2	2	0
Total SASS5 score		69	44	24	83	80	70	27	98	59									
No. of families		9	7	5	13	13	11	6	16	12									
ASPT		7.67	6.29	4.80	6.38	6.15	6.36	4.50	6.13	4.92									
Total IHAS					67				81	81									
IHAS - Habs sampled					27				42	42									
IHAS - Stream condition					40				39	39									
Suitability score		5	5	4	14	7	3	6	16	14									

Key: **Preference**
0 - No preference (does not o
1 - Very low preference Coinc
2 - Low preference
3 - Moderate preference
4 - High preference
5 - Very high preference

Key: High requirement for unmodified water quality
Moderate requirement for unmodified water quality
Low requirement for unmodified water quality
Very low requirement for unmodified water quality
Veg=Vegetation

A = 1-10 individuals; B = 11-100 individuals; C = 101-1000 individuals; ASPT = Average score per taxon.

4.5 Fish/Ichthyofauna

Two earth dams are present in the upper reaches of the Groot Dwars River on the farm De Berg (original study area of proposed DBPNR). Dam 1 (site name De Berg_Dam1) covers approximately 1.0 hectares at full supply (Plate 6). Dam 2 is smaller and covers approximately 0.4 hectares at full supply (Plate 7). Aquatic macrophytes at Dam 1 were dominated by the submerged *Lagarosiphon major*, the floating *Potamogeton nodosus*, and the Bulrush *Typha capensis*. Intensive fish sampling was done in these dams during February 2022 using floating gill nets (mesh size range (mm): 150, 118, 93, 73, 57, 45, 35, 28, 22) and electrofishing by wading in shallow water or from boat. No fish was sampled during the survey (as also observed in 2020/21 surveys) and it can therefore be confirmed that **there is currently no fish present in these dams**. The absence of indigenous fish from these dams (and the upper reaches of the Groot Dwars River) inside DBPNR is thought to be a natural phenomenon as a result of the abundance of natural migration barriers (waterfalls, cascades, large boulders) that occurs within the mountain stream (BMU7) zone of this river. Fish in the Groot Dwars River is therefore primarily limited to the lower reaches (BMU8: Rivers) that falls outside of the DBPNR. It was promising that no alien fish is currently present within these dams since it was suggested by locals that these dams may have been historically stocked with Rainbow Trout (*Oncorhynchus mykiss*). The presence of any alien fish in these dams may have posed a serious threat to the natural indigenous biodiversity of this area as well as the downstream reaches that contains fish species of conservation concern.

Plate 6: De Berg Dam 1 (2022-02).

Plate 7: De Berg Dam2_ (2020-12)

Fish sampling (electrofishing) was also performed at various of the stream and wetlands within the original DBPNR study area on the Groot Dwars River (sites US-Dam1, GD_De Berg, GD_Sterkfontein, GD N-trib), Everest tributary (Everest 1, 2 and 3) and unnamed tributary of the Klip River (sites KR1 and KR2) between 2020 and 2022 (Plates 8 to 10). No fish was present at these sites, further confirming the natural absence of fish from these most upper reaches within the DBPNR. As discussed above the absence of indigenous fish from these reaches may be attributed to the presence of natural migration barriers (waterfalls, cascades), especially in the Groot Dwars River. The potential contribution of man-made migration barriers (in the form of dams and bridges) in limiting natural movement of fish can however not be disregarded (especially in the Everest tributary and potentially also the Klip River system).


Plate 8: Site GD N-trib (2022-02).


Plate 9: Site KR1 (2022-02).

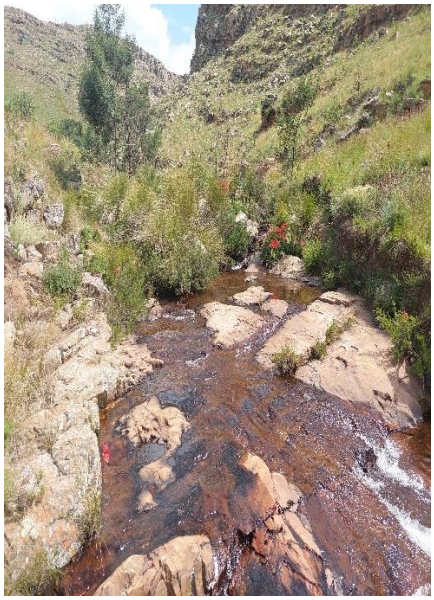
Plate 10: Site Everest 3 (2022-02).

As mention previously in this report a new section, namely the farm Goedehoop, was added to the study area in 2022 after completion of most of the initial DBPNR fieldwork. Limited sampling and visual observations were performed at selected sites of this new section during the end of March 2022. Electrofishing was performed at two dams (sites GH Dam1 and GH Dam2) as well as the Potspruit reach (site PS1) (Plates 11 to 13). The presence of one indigenous fish species, namely *Enteromius cf. anoplus/motobensis* was confirmed at all sites sampled. This species was relatively abundant, especially at site PS1 in the Potspruit (CPUE of 79 individuals/hr). The presence of the alien Rainbow trout (*Oncorhynchus mykiss*) was also confirmed (visual observation) in the larger dam (site GH Dam2) on the property. The fish population of this dam should be further investigated in future through more intensive sampling. The presence of the aggressive predatory Rainbow trout in this dam is a threat to the indigenous fish of the system. It was however promising to note that the indigenous barbs (*E. anoplus/motobensis*) also occurred in the marginal vegetation of this dam, and this species was abundant both upstream (GH Dam1) and downstream (site PS1) at the time of sampling in March 2022.


Plate 11: Site PS1 (2022-03)


Plate 12: Site GH Dam1 (2022-03)


Plate 13: Site GH Dam 2 (2022-03)

5. BOOYSENDAL AQUATIC BIODIVERSITY

5.1 FISH

No previous fish information was available for the DBPNR study area and hence the current status and distribution of fish with this area is based on the surveys conducted on selected sites by Clean Stream Biological Services between 2020 and 2022. These surveys confirmed the absence of fish from all rivers and streams (Groot Dwars River, Everest tributary and Klip River tributary) within the original DBPNR study area (farms De Berg, Triangle and Sterkfontein). The absence of indigenous fish from these upper catchment streams inside DBPNR is thought to be a natural phenomenon as a result of the abundance of natural migration barriers (waterfalls, cascades, large boulders) that occurs within the mountain stream (BMU7) zone. It was also promising that no alien fish species were present in the two dams on the farm De Berg (previously thought to potentially contain alien Rainbow trout).

Limited fish sampling and visual observations performed at selected sites of the new section (farm Goedehoop) during the end of March 2022 confirmed the presence of one indigenous fish species, namely *Enteromius cf. anoplus/motobensis* within the Potspruit river system on this farm. The presence of the alien Rainbow trout (*Oncorhynchus mykiss*) was also confirmed (visual observation) in the larger dam on the property.

The small barb (Plate 14) sampled in the Potspruit on the farm Goedehoop closely resembles *Enteromius motebensis* (the Marico barb) and *Enteromius anoplus* (Chubbyhead barb). The identification of this species remains uncertain and can only be confirmed by further detailed (including genetic) analyses. If this species is not *E. motobensis* or *E. anoplus*, it is likely to be a unique genetic linage of the complex Chubbyhead Barb group of species currently under review in South Africa. It must further also be mentioned that many records currently ascribed to *Enteromius motebensis* and *Enteromius anoplus* in the eastern Lowveld of Mpumalanga may be synonymous with a potential new species *Enteromius sp.nov. "Ohrigstad"* proposed by Engelbrecht & Van Der Bank (1996). A recent study of *E. motebensis* within the Groot Marico catchment found unique haplotypes in two tributaries that required conservation (van der Walt *et al*. 2017). Previous genetic (unpublished) studies of the Enteromius species within the Northam Booysendal study area (Groot Dwars River: BMU8) downstream of DBPNR suggest that this population is genetically unique, as a result of its isolated distribution, and still needs to be described (Dr. Francois Roux, MTPA, *pers. comm.).* The taxonomy of the Enteromius species in the Goedehoop (and greater Northam Booysendal) area remains uncertain and should be confirmed by further studies (that includes genetic analyses).

Until the identification of this species has been clarified, it will be referred to as *Enteromius cf. anoplus/motebensis* and considered to potentially be a fish species of conservation concern. *Enteromius motebensis* is listed by the International Union for Conservation of Nature (IUCN) as near-threatened (NT) [B1b(ii,iii,iv)+2b(ii,iii,iv)](Table 14) while *E. anoplus* is currently listed as Least concern(LC) (IUCN, 2021). Previously all records from the Eastern Lowveld catchments were recognised as *Enteromius anoplus* for the purpose of the IUCN Red List Assessment, accepting that a taxonomic revision of this group is required (Woodford, 2017). The IUCN assessment only considered records from the western Limpopo River Catchment as *Enteromius motebensis*, being listed as Near-Threatened. *Enteromius motebensis* typically occurs in headwater streams where it prefers slow-flowing pools. Headwater stream fish communities are increasingly becoming isolated in headwater refugia as a result of direct and indirect threats further downstream (e.g. predatory fish species such as alien bass and rainbow trout, pollution, flow changes due to abstraction and habitat degradation) (Ellender and Weyl, 2015). This range restriction and isolation gives rise to a high degree of genetic variation and endemism (Abell *et al.* 2007), thus making them vulnerable to extinction. Globally and locally, headwater ecosystems are under increasing threat from human disturbance (van der Walt *et al.* 2017).

Plate 14: *Enteromius cf. anoplus/motobensis* sampled in Potspruit (Goedehoop, 2022-03).

Table 14: Conservation status of fish species of concern.

SCIENTIFIC NAME	IUCN Red list status[1]	TOPS[2]	Notes
Enteromius cf. motebensis *(Enteromius cf. anoplus)*	Near-threatened B1b(ii,iii,iv)+2b(ii,iii,iv)	Not listed	Endemic SA. Complex of genetic unique species & populations *E. motebensis* (North West/Croc East system).

1 – IUCN (2016-3: Version 3.1): LC-Least concern, 2- NEM:BA (10 of 2004): TOPS (RSA Threatened or protected species).

Alien fish species

The presence of one alien invasive fish species (Government Gazette No. 40166: 29 July 2016), namely the Rainbow trout (*Oncorhynchus mykiss*) (Plate 15) was confirmed (visual observation) within the Potspruit system on the farm Goedehoop (newly added section of DBPNR). This aggressive alien predator threatens biodiversity through predation on indigenous fish and invertebrate populations. Ideally it should be aimed to remove or control any alien fish species occurring within nature reserves or area of high biodiversity conservation importance. It was however promising to note that the only indigenous fish species sampled in the Goedehoop section of the study area (*E. cf. anoplus/motebensis*) was abundant both upstream and downstream of the dam that housed the trout, and the indigenous fish was also present along the edges of the dam where the trout occurred. It therefore seems that the trout is not currently an immediate threat to the occurrence of this indigenous fish in the upper Potspruit system. Should the land owners decide to maintain the Rainbow Trout population within this dam, the status of the indigenous fish should be monitored closely.

Plate 15: Rainbow trout (*Oncorhynchus mykiss*)

5.2 AQUATIC MACRO-INVERTEBRATES

Aquatic macroinvertebrate diversity of the study area was primarily assessed on family level through the application of the South African Scoring System (version 5) (SASS5) protocol. A once-off survey was also performed by Dr. R. Palmer to collect primary data on aquatic macroinvertebrate species at representative sites.

Species of Conservation Concern

Distribution maps provided by the IUCN Redlist (https://www.iucnredlist.org/) indicate the potential presence of one species of aquatic macroinvertebrate of conservation concern that could be expected within the DBPNR study area, namely, *Pseudagrion newtoni* (VU: Vulnerable). *Pseudagrion newtoni (*common name is the Harlequin sprite) is a species of damselfly in the family Coenagrionidae. The presence of this species was not confirmed during the current survey but there is a high probability that this species may be present within the DBPNR. The field survey for this report found no threatened aquatic macroinvertebrate species in the Study Area. However, the following rare, endemic or range-restricted taxa were recorded:

- *Mesostoma sp.* (Typhloplanidae)
- *Afronemoura stuckenbergi* (Notonemouridae)
- *Demoreptus cf. monticola* (Baetidae)
- *Elporia marieps* (Blepariceridae)
- *Simulium debegene* (Simuliidae)

Invertebrate species composition

The most diverse group were true flies (21 taxa), beetles (9 taxa) and bugs (6 taxa). The diversity of mayflies was low (5 taxa) and included the most hardy and widespread of all mayflies in southern Africa, *Baetis harrisoni.* One species of crab was recorded, namely the Natal river crab *Potamonautes sidneyi*. This species is the most widespread of all crabs in Southern Africa (Hart *et al.* 2001). Aquatic snails were absent, although they are expected to be presented in the lower-lying portions of the study area. Functional feeding in all zones was dominated by shredders, except downstream of De Berg Dam 1 (Site A3), where there was a high abundance of filterers. The change in functional feeding downstream of the dam is attributed to the release of plankton from the dam.

Invertebrate families (SASS5)

A total of forty-seven (47) macroinvertebrate families were sampled in the valley- bottom wetlands and seeps (BMU5) and mountain streams (BMU7) within the DBPNR study area between 2020 and 2022 (Table 15). This reflects a relatively high diversity of aquatic macroinvertebrate families and is a reflection of highly diverse aquatic habitats as well as areas with very good water quality. Limited sampling (one site) within BMU 5 (wetlands and seeps) indicated the presence of 18 macroinvertebrate taxa (Table 16), while 47 taxa was sampled within BMU 7 (Mountain streams).

Five taxa/groups with a high requirement for unmodified water quality was sampled in the study area, namely *Blepharoceridae* (Net-winged midges) (Plate 16), *Notonemouridae* (Stoneflies) (Plate 17), *Perlidae* (Stoneflies) (Plate 18), >2spp. *Baetidae* (Small minnow flies) and *Pyralidae* (Aquatic caterpillars) (Table 16). A further eleven (11) taxa with a moderate requirement for unmodified water quality (such as *Psephenidae*-Water pennies (Plate 19) was also sampled in the study area (Table 16). The presence of these intolerant taxa at specific sites indicates excellent water quality prevailing at present in most reaches of the DBPNR study area. Special mention must be made of the presence of *Blepharoceridae* (Net-winged midges) (Plate 16) sampled at various sites in the upper reaches of the Groot Dwars River and Everest tributary (BMU7': Mountain stream). This taxon attains the highest possible SASS5 sensitivity score/intolerance rating of 15, indicating that it is only found in the most pristine sites with unmodified water and habitat quality (SASS5 taxa are rated from 1 for the most tolerant taxa to 15 for the most intolerant taxa). The presence is a certain indication that the upper Groot Dwars River catchment (including upper Everest tributary) falling mostly within the DBPNR is currently maintaining very good water quality and play an important role in sustaining this river

in a good ecological condition. It is therefore strongly recommended that these areas should be conserved and no activities should be allowed that may jeopardise the water quality and overall ecological integrity of these source streams.

The macroinvertebrate taxa sampled in the study area vary in their requirement for flow/velocities as well as cover features (Table 15). It is therefore essential to maintain a diversity of habitats, together with good water quality, in an attempt to conserve the aquatic macroinvertebrate diversity of the DBPNR. Most of the taxa with a high and moderate requirement for unmodified water quality generally prefers fast flowing water (>0.3 m/s) with cobble as substrate, a general feature of the mountain streams (BMU7) (Table 15). It therefore furthermore emphasises the importance of maintaining good flow (high velocities) (no damming, limit abstraction and evaporation), good water quality (prevent pollution) and clear stone habitat (prevent erosion, sedimentation) in an attempt to conserve these intolerant taxa and overall diversity of the study area.

Plate 16: *Blepharoceridae* (Net-winged midges) (Dr. R. Palmer)

Plate 17: Notonemouridae (Stoneflies) (Dr. R. Palmer)

Plate 18: *Perlidae* (Stoneflies)

Plate 19: *Psephenidae* (Water penny beetles)

Table 15: Aquatic macroinvertebrate taxa (families) sampled in the different BMU's of the DBPNR study area (2020 to 2022) indicating their requirement for unmodified water quality, flow and cover preferences.

Taxon	Common name	BMU5	BMU7	Flow (in m/s) preference				Cover preference				
		Wetlands & seeps	Mountain streams	<0.1	0.1-0.3	0.3-0.6	>0.6	BEDROCK	COBBLES	VEG	GSM	WATER COLUMN
Blepharoceridae	Net-winged midges	-	X	0	0	3	4	2	3	0	0	0
Notonemouridae	Stoneflies	X	X	1	1	2	4	1	4	1	0	0
Perlidae	Stoneflies	-	X	1	1	1	5	1	4	1	0	0
Baetidae > 2 spp.	Small minnow flies	-	X									
Pyralidae	Aquatic caterpillars	-	X	1	1	3	2	0	2	3	0	0
Platycnemidae	Damselflies	-	X	2	3	1	1	0	2	3	0	0
Philopotamidae	Caseless caddisflies	-	X	0	1	2	3	1	4	1	1	0
Psephenidae	Water penny beetles	-	X	0	1	3	4	1	4	1	0	0
Athericidae	Snipe flies	-	X	0	1	2	2	1	4	1	1	0
Leptophlebiidae	Prongills	X	X	3	2	2	1	1	3	2	0	0
Tricorythidae	Stout crawlers	-	X	0	1	1	4	1	4	1	0	0
HYDRACARINA	Water mites	-	X	0	2	2	0	1	1	2	3	1
Chlorolestidae	Damselflies	X	X	3	2	1	0	0	1	4	0	0
Lestidae	Damselflies	-	X	4	1	0	0	0	1	4	1	0
Aeshnidae	Dragonflies	X	X	1	2	2	2	0	3	2	0	0
Ecnomidae	Caseless caddisflies	-	X	1	5	0	0	2	3	2	0	0
Naucoridae*	Creeping water bugs	-	X	2	2	3	0	1	1	1	1	4
Baetidae 2 spp.	Small minnow flies	X	X									
Caenidae	Cainflies	-	X	3	2	1	1	0	2	1	3	0
Gomphidae	Dragonflies	-	X	0	2	3	0	0	1	0	5	0
Hydropsychidae 2 spp.	Caseless caddisflies	-	X									
Hydroptilidae	Micro caddisflies	-	X	0	3	2	2	1	2	3	1	0
Leptoceridae	Cased caddisflies	X	X	0	1	3	2	2	2	2	2	0
Ancylidae	Limpets	-	X	1	2	2	1	3	2	1	0	0
Gerridae*	Pond skater	X	X	4	1	0	0	0	0	0	0	5
Veliidae*	Broad-shouldered water strid	X	X	5	1	1	0	0	0	0	0	5
Dytiscidae (adults*)	Predacious diving beetles	X	X	4	2	1	0	1	2	3	1	2
Gyrinidae (adults*)	Whirligig beetles	X	X	1	2	2	3	0	0	0	0	5
Hydrophilidae (adults*)	Water scavenger beetles	-	X	0	2	2	0	0	0	3	2	2
Ceratopogonidae	Biting midges	X	X	2	2	2	4	2	3	2	2	0
Simuliidae	Black flies	X	X	0	2	2	4	2	3	2	0	0
Tabanidae	Horseflies	-	X	2	3	1	0	0	2	0	3	0
Tipulidae	Crane flies	-	X	3	4	1	1	1	2	0	3	0
Baetidae 1 sp.	Small minnow flies	X	X	2	2	2	2	2	2	2	2	1
Coenagrionidae	Damselflies	X	X	1	2	3	1	0	1	4	1	0
Libelludae	Dragonflies	-	X	1	2	3	1	1	4	0	1	0
Hydropsychidae 1sp.	Caseless caddisflies	-	X	0	1	2	4	2	3	1	0	0
TURBELLARIA	Flatworms	X	X	1	2	3	4	1	4	0	0	0
Leeches	Leaches	-	X	2	2	1	1	0	4	1	1	0
Potamonautidae*	Crabs	-	X	1	1	3	2	0	3	1	1	0
Corixidae*	Water boatmen	-	X	2	3	1	0	1	1	1	1	4
Nepidae*	Water scorpions	X	X	4	1	0	0	0	0	5	0	0
Notonectidae*	Back swimmers	X	X	4	1	0	0	0	0	2	0	4
Chironomidae	Midges	X	X	1	3	2	2	2	2	2	2	0
Oligochaeta	Aquatic earthworms	-	X	2	2	2	1	0	1	0	4	0
Culicidae*	Mosquitoes	X	X	3	1	0	0	0	0	0	0	5
Muscidae	House flies	-	X	4	2	2	0	1	1	1	1	4

High requirement for unmodified water quality
Moderate requirement for unmodified water quality
Low requirement for unmodified water quality
Very low requirement for unmodified water quality

Key: Preference
0 - No preference (does not occur)
1 - Very low preference Coincidental
2 - Low preference
3 - Moderate preference
4 - High preference
5 - Very high preference

5.3 DIATOMS

Diatoms are of great ecological importance because of their role as primary producers, and they form the base of the aquatic food web. Diatoms have been shown to be reliable indicators of specific water quality problems such as organic pollution, eutrophication, acidification and metal pollution, as well as for general water quality. This information however also provides preliminary data on the diversity of diatom species in an area. Representative sites were sampled within the proposed DBPNR study area to gain some baseline diatom results of the area during the February 2022 aquatic survey.

Fifty-nine (59) diatom species were identified (first 400 counted per sample) at the five sampling sites assessed in DBPNR during February 2022 (Table 17). Four of the five sites in the proposed De Berg Nature Reserve were characterised by **high biological water quality** reflecting near pristine conditions, while the remaining site was rated as having moderate biological water quality (Site GD N-trib). Endemic species with a preference for high biological water quality were observed at Sites KR1, Everest 2 and 3, and Site GD N-tib. Endemic species occurred at high abundance at Sites KR1, Everest 2 and 3 (Table 16). These species, based on the experience of the diatomologist, are scarce and have only been observed in the upper reaches of high altitude streams or the upper reaches near the origins of streams where anthropogenic activity is limited.

Table 16: List of diatom species collected during February 2022 (Endemic species are shaded).

Species	US DAM 1	KR1	EVEREST 2	EVEREST 3	GD NTRIB
Achnanthes standeri Cholnoky		175	166	162	
Achnanthes subaffinis Cholnoky			61	29	
Achnanthes subsaxonica Cholnoky			88	42	10
Achnanthidium crassum (Hustedt) Potapova & Ponader	10	8			
ACHNANTHIDIUM F.T. Kützing	7				
Achnanthidium macrocephalum (Hustedt) Round & Bukhtiyarova	2				3
Achnanthidium minutissimum (Kützing) Czarnecki	9	16	37	26	102
Achnanthidium subatomoides (Hustedt) Monnier, Lange-Bertalot et	1				
ADLAFIA Moser Lange-Bertalot & Metzeltin					3
AULACOSEIRA G.H.K. Thwaites				14	
Brachysira brebissonii Ross in Hartley	1			3	1
Brachysira neoexilis Lange-Bertalot	1				38
Caloneis bacillum (Grunow) Cleve					2
Chamaepinnularia mediocris (Krasske) Lange-Bertalot	2				1
Cymbella naviculiformis Auerswald					3
CYMBELLA C.Agardh	6				
Encyonema minutum (Hilse) D.G. Mann				5	
Encyonema theronii (Cholnoky) Krammer		168	4	18	1
Encyonopsis leei var. *sinensis* Metzeltin & Krammer			1	1	
Encyonopsis microcephala (Grunow) Krammer			2		
Encyonopsis microcephala var. *robusta* (Hustedt) Krammer	3				
EUNOTIA C.G. Ehrenberg	2				
Eunotia bilunaris (Ehr.) Mills	112				14
Eunotia exigua (Brebisson ex Kützing) Rabenhorst	11	6		4	1
Eunotia flexuosa (Brebisson) Kützing	3				1
Eunotia hugenottarum Cholnoky				12	3
Eunotia incisa Gregory	1				2
Eunotia minor (Kützing) Grunow	26		1	4	36

Species	US DAM 1	KR1	EVEREST 2	EVEREST 3	GD NTRIB
Eunotia muscicola Krasske	5				
Eunotia paludosa Grunow	4				
Eunotia rhomboidea Hustedt	21			2	2
FRUSTULIA L. Rabenhorst	2				
Frustulia crassinervia (Brebison) Lange-Bertalot et Krammer	6				
Frustulia rhomboides (Ehrenberg) De Toni					3
Frustulia rhomboides var. *amphipleuroides* (Grunow) De Toni	1				
Frustulia saxonica Rabenhorst	1				
Frustulia vulgaris (Thwaites) De Toni	7				5
GOMPHONEMA C.G. Ehrenberg	2		37	14	
Gomphonema acidoclinatum Lange-Bertalot & Reichardt					1
Gomphonema acuminatum Ehrenberg					2
Gomphonema affine Kützing					6
Gomphonema angustatum (Kützing) Rabenhorst					3
Gomphonema gracile Ehrenberg					11
Gomphonema lagenula Kützing	3				100
Gomphonema parvulum (Kützing)				1	
NAVICULA J.B.M. Bory de St. Vincent	1				1
Navicula angusta Grunow		2			
Navicula arvensis var. *maior* Lange-Bertalot					1
Navicula veneta Kützing					1
NITZSCHIA A.H. Hassall	1			1	9
NUPELA W. Vyverman & P. Compere				60	
PINNULARIA C.G. Ehrenberg	1				
Rhopalodia operculata (Agardh) Hakånsson					2
Sellaphora seminulum (Grunow) D.G. Mann					1
STAURONEIS C.G. Ehrenberg					1
Stenopterobia delicatissima (Lewis) Brebisson ex Van Heurck	1	21	2	2	2
Synedra rumpens Kützing	1		1		9
Tabellaria flocculosa (Roth) Kützing	146	4			2
Ulnaria biceps (Kützing) Compere					17
Total count	**400**	**400**	**400**	**400**	**400**

6. SUMMARISED AQUATIC BIODIVERSITY (PER BMU)

BMU 5: Valley-bottom wetlands and seeps (Plate 20)

BMU5 (Valley bottom wetlands and seeps include the hygrophytic and hydrophytic grass and sedge dominated wetland communities on hydromorphic soils of channeled and unchanneled valley-bottom wetlands and seeps. Most seeps are connected to valley-bottom wetlands but a few are directly connected to mountain streams. All of the streams comprising this BMU form part of the most upper catchments (source zones) of the Groot Dwars River and their tributaries (including Everest Stream), Klip River tributary and Potspruit. These catchments are classified as Critical Biodiversity Areas or Ecological Support Areas (MBSP, 2014) and the Groot Dwars River (SQ reach B41G-721) within this BMU are also classified as a National Freshwater Ecosystem Priority Area (NFEPA's). The seeps were characterised by peat soils and *Sphagnum* moss and aquatic biotopes in this zone comprised mostly shallow seeps that are likely to be active seasonally, and smaller areas that are likely to have permanent surface water. The source zones comprised springs, bedrock sheets, pools and shallow runs.

The biodiversity in terms of true aquatic faun of this zone is limited due to the limited availability of a water column. The current study confirmed that fish was absent from this BMU as a result of the natural habitat limitation as well as the various natural migration barriers (waterfalls, cascades) preventing movement of fish into these reaches. Limited sampling confirmed the presence of various algae and moss species and at least 18 macroinvertebrate families (that includes a many species). Diatom analyses confirmed that some sections of this BMU can be classified within an ecological category A and that biological water quality was excellent. These source zones play a critical role in sustaining the mountain streams (BMU7) and lower river reaches and no activities should be allowed that may jeopardise the water quality and overall ecological integrity of these source zones. This BMU is considered to be of **high** biodiversity conservation value in terms of aquatic biodiversity.


Plate 20: BMU5: Valley-bottom wetlands and seeps.

BMU 7: Mountain Streams (Plate 21)

BMU7 (Mountain streams) includes the perennial and non-perennial mountain streams (mostly 1st and 2nd order streams) which occur on both igneous (mostly norite) and sedimentary (sandstone) geology. In terms of geomorphological zone classification ((Rowntree & Wadeson, 1999), BMU 7 comprises entirely of watercourse reaches categorized as mountain streams (zone B) and Mountain headwater streams (zone A). Aquatic biotopes in this zone comprised bedrock, pools, runs, riffles, cascades and waterfalls. All of the streams comprising this BMU form part of the most upper catchments of the Groot Dwars River and their tributaries (including Everest Stream), Klip River tributary and very small stretches of the Potspruit on the farm Goedehoop. Many reaches of this unit are classified as Critical Biodiversity Areas or Ecological Support Areas (MBSP, 2014) and the Groot Dwars River (SQ reach B41G-721) within this BMU are also classified as a National Freshwater Ecosystem Priority Area (NFEPA's).

The current study furthermore indicated that fish was absent from the most upper reaches of the mountain streams of the Groot-Dwars River (De Berg and Sterkfontein Portion3), Everest Tributary (on De Berg) and

Klip River tributary within the original DBPNR study area. It is estimated that the absence of fish from these areas may be a natural phenomenon as a result of various natural migration barriers (waterfalls, cascades) within these high gradient river reaches. One indigenous fish species (*E. cf. anoplus/motebensis*) and one alien fish species (*O. mykiss*) was confirmed from the Potspruit section on the farm Goedehoop (new section of DBPNR). The identification of E. cf. anoplus/motobensis requires further verification but currently it should be viewed as a potential species of conservation concern due to the fact that E. motebensis is currently listed as Near-Threatened (NT) by the IUCN. The current study furthermore confirmed the presence a highly diverse aquatic macroinvertebrate richness with 47 macroinvertebrate families (that includes a high number of species) recently (2020 to 20220) sampled in this BMU within DBPNR. The presence is a various taxa with a high (5 taxa) and moderate (11 taxa) requirement for unmodified water quality indicate that many of the mountain stream reaches in the study area is currently still in an excellent ecological condition. This was also confirmed by diatom analyses that indicate excellent (ecological category A) biological water quality prevailing in many streams within DBPNR (many endemic diatom species with a preference for high biological water quality were also confirmed). These upper catchment streams also play an important role in sustaining the lower river reaches (adequate flow and water quality through dilution of pollutants) and it is therefore strongly recommended that these mountain stream catchment areas (entire DBPNR) should be conserved and no activities should be allowed that may jeopardise the water quality and overall ecological integrity of these source streams. This BMU is considered to be of **very high** biodiversity conservation value in terms of aquatic biodiversity.

BMU7: MOUNTAIN STREAMS

Plate 21a: Most Upper Groot Dwars River in De Berg (site GD_DeBerg)

Plate 21b: Most upper Groot Dwars River (site GD_SterkfonteinP3)

Plate 21c: Tributary of Groot Dwars River (site GD_N-trib)

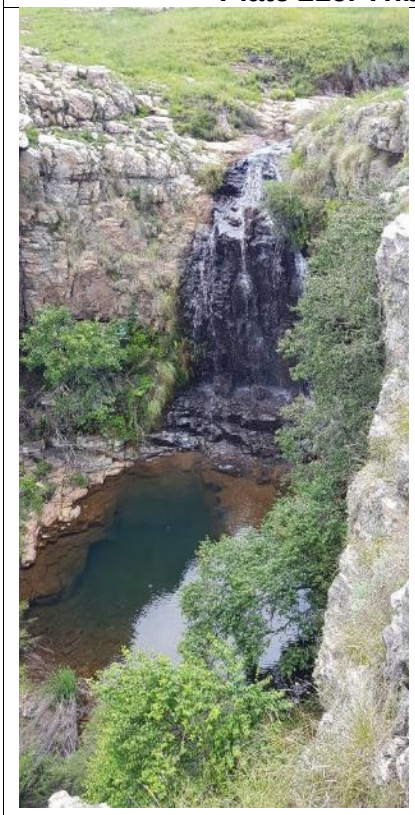

Plate 21d: Most upper reaches of Everest Tributary inside De Berg (Site Everest 1&2)

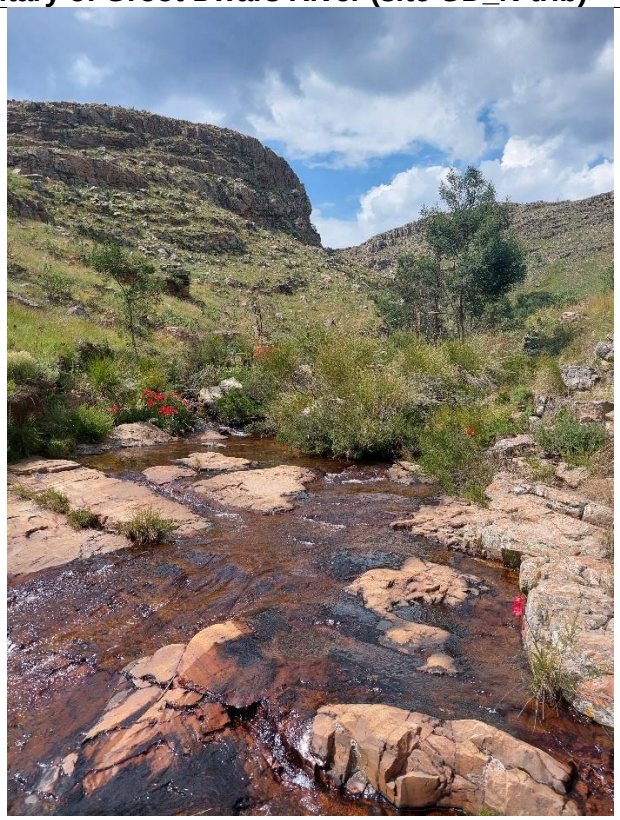

Plate 21e: Everest Tributary where it leaved DBPNR (Site Everest 3)

BMU 11: Dams (Plate 22)

BMU11 (Dams) includes the artificial wetlands created by various relatively small, earth-walled farm dams (such as De Berg Dams 1 and 2 and Goedehoop dams 1 and 2). According to the wetland classification system of (Ollis *et al*. 2013), all of the farm dams are 'in –channel' earth-walled dams. BMU11 (dams) are artificially created aquatic habitats that transformed natural river reaches (mostly lotic ecosystems) into stagnant (lentic ecosystems). Although these dams created artificial habitats that are utilised by various aquatic fauna, they should be viewed as an impact/threat to the natural biodiversity of the study area. These

dams often create migration barriers to the natural movement of fish and create suitable habitat for some (mostly unwanted) fish species, including alien fish. The dams furthermore also alter the natural aquatic biota composition in the rivers directly downstream of the dams due to flow and water quality changes. The current study indicated that functional feeding of macroinvertebrates at all sites surveyed was dominated by shredders, except downstream De Berg Dam 1, where there was a high abundance of filterers. The change in functional feeding downstream of the dam is attributed to the release of plankton from the dam. Seepage downstream of the dam was also characterised by dense growth of the protobacterium *Leptothrix ochracea* which clogged interstitial spaces and created conditions that were unsuitable for aquatic macroinvertebrates. This species is typically associated with oxidation of iron. The current study focused on the natural (untransformed) habitats with potentially high biodiversity conservation potential in the study area with limited sampling performed in the dams as part of this study.

Fish sampling in the dams on the farm De Berg (DB Dam 1 and 2) confirmed that **there is currently no fish present in these dams**. The absence of indigenous fish from these dams (and the upper reaches of the Groot Dwars River) inside DBPNR is thought to be a natural phenomenon as a result of the abundance of natural migration barriers (waterfalls, cascades, large boulders) that occurs within the mountain stream (BMU7) zone of this river. It was promising that no alien fish is currently present within these dams since it was suggested by locals that these dams may have been historically stocked with Rainbow Trout (*Oncorhynchus mykiss*). The presence of any alien fish in these dams may have posed a serious threat to the natural indigenous biodiversity of this area as well as the downstream reaches that contains fish species of conservation concern. Limited fish sampling and visual observations confirmed the presence of one indigenous fish species, namely *Enteromius cf. anoplus/motobensis* and the alien Rainbow trout (*Oncorhynchus mykiss*) in the dams on the farm Goedehoop (site GH Dam 1 and 2). The presence of the aggressive predatory Rainbow trout is a threat to the indigenous fish of the system. It was however promising to note that the indigenous barb (*E. anoplus/motobensis*) also occurred in the marginal vegetation of this dam, and this species was abundant both upstream (GH Dam1) and downstream (site PS1) at the time of sampling in March 2022. It therefore seems that the trout is not currently an immediate threat to the occurrence of this indigenous fish in the upper Potspruit system. Should the land owners decide to maintain the Rainbow Trout population within this dam, the status of the indigenous fish should be monitored closely. Limited aquatic macroinvertebrate sampling performed on De Berg Dam 1 indicated a moderate diversity of macroinvertebrates, with 17 taxa recorded in around the dam. The dam supported taxa that are typically associated with standing water, such as *Chironomidae, Ceratopogonidae, Gerridae, Naucoridae and Pleidae*. High numbers of adult Swamp bluet *Africallagma glaucum* (*Coenagrionidae*) were recorded around the dam. The land owner should aim to limit the construction of any additional instream dam walls and also remove any redundant dams from the river systems under their control.

Plate 22a: Dam in most upper Groot Dwars River on De Berg (Site De Berg_Dam1)

Plate 22b: Dam in most upper Groot Dwars River on De Berg (Site De Berg_Dam2)

Plate 22c: Site GH Dam 1 on farm Goedehoop (Potspruit)

7. BIODIVERSITY IMPACTS/THREATS AND MANAGEMENT RECOMMENDATION (AQUATIC BIODIVERSITY)

The current section provides an overview of potential impacts, threats or risks to the aquatic biodiversity within the DBPNR study area (Table 17). The potential impacts are described, an indication provided of the relevance to specific BMU's as well as the species or aspects of concern regarding the specific impact. Recommendations are also made regarding potential management and mitigations measures that can be considered for implementation. The aim of the current report is not to replace any previous impact assessments and recommended management actions, which should still be utilised and implemented where applicable.

Table 17: Broad overview of potential impacts and threats to aquatic biodiversity of the study area, relevant BMU's and species/aspects of concern, as well as recommended management actions.

IMPACT/THREAT	DESCRIPTION OF IMPACT/THREAT	RELEVANCE TO BMU's	SPECIES/ASPECTS OF CONCERN	RECOMMENDED MANAGEMENT ACTIONS
Impact 1: Altered hydrological regimes (increased or decreased flows/water levels)	Dams (BMU11), weirs, bridges, pipeline crossings: These activities may result in flow alteration (storage/pooling of water, delay in floods/flushes/freshets, increased evaporation) and often abstraction (loss of water from system). The construction of buildings and roads also alter natural drainage patterns Excavations/trenches/canals may cut-off of or alter surface flow and underground seepage. DBPNR specific impacts: De Berg Dams 1 and 2 and Goedehoop Dams 1 and 2.	5, 7	All aquatic biota will be impacted by altered flow regime, but especially flow intolerant invertebrates.	Determine flow requirements and comply with ecological reserve (quality and quantity) Remove any redundant dams, bridges and rehabilitate these areas (removal should be done through formal process to minimise any potential environmental impacts, such as increased sedimentation of downstream reaches, spreading of alien fish species, etc.). Prevent the construction of additional in-stream dams/bridges/pipelines across aquatic ecosystems. No development should occur within the 1: 100-year flood line of any watercourse. Make sure that the natural flow of all drainage lines is kept intact and prevent erosion at all cost. It is important that the natural diversity of habitats must be maintained to cater for the diversity of aquatic fauna in the study area.

IMPACT/THREAT	DESCRIPTION OF IMPACT/THREAT	RELEVANCE TO BMU's	SPECIES/ASPECTS OF CONCERN	RECOMMENDED MANAGEMENT ACTIONS
Impact 2: Fragmentation / migration barriers	Dam walls as well as poorly constructed bridges and pipeline crossings create migration barriers that hinder the free movement of fish. Pollution/release of water of poor quality may cause "chemical migration barriers", especially to intolerant aquatic biota. Due to the absence of fish from the most upper reaches (mountain streams) the dams in these areas will have no/minimal migration impacts. Goedehoop dam 2 limited and not significant impact on movement of *E. cf. anoplus/motebensis*.	5, 7	Most fish species require free movement within or between reaches (includes NT[3] *E. cf. anoplus/motebensis* to complete life-cycle.	Refrain from building any further in-stream dams, weirs, river crossings. Remove redundant dams/structures to restore the natural river continuum. No dams or weirs, other than those specifically designed for erosion control, may be constructed in wetlands. Unnecessary damming of the river, tributaries, wetlands and seepages should not be allowed. Prevent any pollution/areas of poor water quality as to not create chemical migration barriers.
Impact 3: Water quality deterioration	Alterations to **water quality** (such as eutrophication, increased salinity, increased turbidity) through effluents, storm water runoff, and seepage into streams. Reduced water quality related to potential seepage from infrastructure (such as offices, accommodation) which could have impacts on the aquatic biota. Flushed-out pesticides, detergents, and other poisonous substances.	5, 7	Most aquatic fauna but especially water quality intolerant species/taxa (including NT *E. cf. anoplus/motebensis*).	Prevent surface, ground water, or effluents with poor quality from entering the aquatic ecosystems. Oil and other hydrocarbons must be strictly controlled (limit storage of fuels on site, no servicing/repairs of vehicles, etc.). Implement water quality monitoring program on DBPNR and ensure compliance to water quality guidelines. Implement aquatic biomonitoring programme to monitor any changes in the ecological integrity of the aquatic ecosystem. Identify potential areas where seepage and spills can occur into the natural environment and take preventative measures (such as from infrastructure, sewage treatment facilities). Determine and comply with ecological reserve (quality and quantity). If pesticides or herbicides are used (such as during alien plant control, weed control, firebreaks), products should be chosen responsibly to act in accordance with the sensitive environment and associated ecology. Storage, administration and disposal must be done according to the prescribed methods. Care should

[3] Near-threatened (IUCN)

IMPACT/THREAT	DESCRIPTION OF IMPACT/THREAT	RELEVANCE TO BMU's	SPECIES/ASPECTS OF CONCERN	RECOMMENDED MANAGEMENT ACTIONS
				be taken to prevent any of the pollution from ending up in the wetlands or river. Educate farmers about the importance of invertebrate conservation and encourage use of integrated pest management (IPM) strategies with reduced reliance on pesticides. No dumping of wet or dry material and, in particular, no waste disposal of any kind may be permitted in or near a wetland or stream.
Impact 4: Aquatic habitat deterioration	**Increased siltation/embeddedness of bottom substrates and loss of depth in pools** as a result of sediment inflow. This is the end product of catchment erosion (human activities responsible for removal of vegetation, such as clearing for infrastructure, roads, etc.). Also includes grading of fire breaks, grading of roads through untransformed land and uprooting of alien vegetation. **Removal of riverine vegetation**: The structural habitat of aquatic systems can be significantly degraded by alteration of the riparian zones. **Accelerated** flows downstream of outlets, bridges, canals cause erosion, scouring banks and reducing the availability of marginal vegetation habitats. Erosion will also **increase the turbidity** of the water, affecting species with a high requirement for clear water. Where sediments settle out (sedimentation), substrates will be altered, affecting those species that prefer clear, cobbled substrates. Pool depth will also be reduced, affecting species that prefer deep pools. Bridges, dams, river crossings cause pooling upstream. The inundation upstream of the bridge may also create favourable habitats for unfavourable species and	5, 7	All aquatic species.	Limit surface soil disturbance and manage erosion (especially dirt roads and previously disturbed areas). No development or disturbance should occur within the 1:100-year flood line of any drainage line (including perennial and non-perennial streams) in accordance with the National Water Act (no. 36 of 1998). Demarcate all wetland and riverine boundaries and associated buffer zones No dumping of waste or any other materials is allowed within or close proximity to aquatic ecosystems. Implement all possible erosion control measures. Ensure adequate storm water drainage (infrastructure, roads). Specialist aquatic assessments should be conducted before, and monitoring conducted after disturbance of riverine habitats. It is important to maintain good vegetative cover (overhanging vegetation, undercut banks and substrates). Do not allow removal of vegetation along banks, erosion and alien vegetation encroachment, or sedimentation of rocky substrates. Promote sustainable use of vegetation by local community. Carrying capacity should not be exceeded (conduct grazing capacity assessment implements veld management plan). Trampling at watering areas should be minimized. No activities are to infringe upon the wetland and riverine boundaries or associated buffer zones. Should it be absolutely unavoidable that activities occur within these areas, relevant authorisation should be obtained according to the National

IMPACT/THREAT	DESCRIPTION OF IMPACT/THREAT	RELEVANCE TO BMU's	SPECIES/ASPECTS OF CONCERN	RECOMMENDED MANAGEMENT ACTIONS
	change the overall fish assemblage of this area. BDPNR: The spillway of De Berg Dam 1 is eroding and it fail in the near future. Failure of the Dam would have detrimental ecological impacts on the downstream Groot-Dwars River because of elevated sediments. The upper reaches of the Groot-Dwars River supports range-restricted aquatic biota that are sensitive to elevated sediments. Seepage downstream of De Berg Dam 1 was characterised by dense growth of the protobacterium *Leptothrix ochracea* This bacterium clogged interstitial spaces and created conditions that were unsuitable for aquatic macroinvertebrates. This species is typically associated with oxidation of iron. Spillage from the dam also created conditions suitable for filter-feeding macroinvertebrates, particularly the blackfly *Simulium medusaeforme* and therefore had a measurable impact on the ecological functioning of the Dwars River directly downstream of the dam. Active bank and rill erosion was observed in some areas on DBPNR (such as Everest tributary catchment).			Environmental Management Act (NEMA) 107 of 1998 and Section 21 c and i of the National Water Act 36 of 1998.
Impact 6: Invasion by alien plants (especially in riparian zones)	Results in **decreased water levels** (see impact 1: altered hydrological regime). **Compete with indigenous** riparian plant species, altering natural marginal zone vegetation as cover for aquatic fauna. **Floating alien/exotic vegetation** prevent sunlight from penetrating into the water column, thus interfering with photosynthesizing algae in the water	5,7	Most aquatic species but especially species with preference for marginal vegetation as cover.	Implement an alien plant control programme (conducted as part of current BMP study). Alien plant removal should be emphasised in the natural biotopes. Promote use of alien trees by local communities for fire wood and construction activities.

IMPACT/THREAT	DESCRIPTION OF IMPACT/THREAT	RELEVANCE TO BMU's	SPECIES/ASPECTS OF CONCERN	RECOMMENDED MANAGEMENT ACTIONS
	column, which could lead to oxygen depletion in the water.			
Impact 7: Presence of exotic/alien fish	Presence of exotic fish species impact on indigenous fish through predation, disturbance of bottom substrates, competition for food and habitat, transfer of parasites. Dams (BMU11) especially create artificial habitats for proliferation of alien species. Presence of alien Rainbow trout in Goedehoop Dam is a potential threat to conservation of NT. *E. cf.anoplus/ motebensis.*	7, 8	Most aquatic species but especially small species (such as NT *E. cf. anoplus/motebensis*) is preyed upon by predatory alien fish.	Prohibit stocking of exotic fish and invertebrate species or translocation of indigenous fish species in any dams (BMU11) or rivers within the study area. Educate surrounding farmers/landowners about the threat of alien species. Promote conservation of indigenous species and removal of alien species The presence of the alien predatory Rainbow trout in GH Dam 1 is a potential threat to the indigenous fish of the Potspruit system (farm Goedehoop). It was however promising to note that the indigenous barb (*E. anoplus/motobensis*) also occurred in the marginal vegetation of GH Dam 2, and this species was abundant both upstream (GH Dam1) and downstream (site PS1) at the time of sampling in March 2022. It therefore seems that the trout is not currently an immediate threat to the occurrence of this indigenous fish in the upper Potspruit system. Should the land owners decide to maintain the Rainbow Trout population within this dam, the status of the indigenous fish should be monitored closely through an aquatic biomonitoring programme.
Impact 8: Poaching	Using of destructive methods such as gill nets, piscicides or fish traps can seriously impact on the fish population of the area (reduced abundance and even loss of species).	7, 11	All indigenous fish species.	No poaching was observed or are known to occur currently within DBPNR study area. This is predominantly prevented by access control and patrols by security and should be maintained. Dams (BMU11) are often targeted by poachers and should especially be monitored for any signs of poaching activity.

8. CONCLUSION AND RECOMMENDATIONS

The study area falls within the Olifants (B) Water Management Area (WMA) and specifically quaternary catchments B41G (Groot Dwars River and Everest tributary), B41C (Klip River) and B42F (Potspruit). The current study concluded that the DBPNR study area contains areas of high to very high aquatic biodiversity conservation importance. The present ecological status of most of the aquatic ecosystems falling within the DBPNR study area is largely natural to slightly modified (ecological category A to B) with high to very high ecological importance and sensitivity. The Groot Dwars River reaches within the study area is furthermore classified as a National Freshwater Ecosystem Priority Areas (NFEPA's) which elevates their conservation importance. The Groot-Dwars River (sub-quaternary reach B41G-00721) is considered by the Mpumalanga Biodiversity Conservation Plan to be a "Critical Biodiversity Areas" (FEPA River), while the various tributaries draining these sub-catchments (Everest Tributary, etc.) are classified as "Ecological Support Areas: Important sub-catchments" (FEPA sub-catchments). The National Environmental Screening Tool indicated that the aquatic biodiversity sensitivity of the majority of the DBPNR study area was *Very High.* The two most important aquatic biodiversity management units of concern within the DBPNR study area are BMU 5: Valley-bottom wetlands and seeps and BMU 7: Mountain Streams, while limited artificially created systems (BMU 11: Dams) are also present.

The current study confirmed the absence of fish from all rivers and streams (Groot Dwars River, Everest tributary and Klip River tributary) within the original DBPNR study area (farms De Berg, Triangle and Sterkfontein). The absence of indigenous fish from these upper catchment streams inside DBPNR is thought to be a natural phenomenon as a result of the abundance of natural migration barriers (waterfalls, cascades, large boulders) that occurs within the mountain stream (BMU7) zone. It was also promising that no alien fish species were present in the two dams on the farm De Berg (previously thought to potentially contain alien Rainbow trout). Limited fish sampling and visual observations performed at selected sites of the new section (farm Goedehoop) confirmed the presence of one indigenous fish species, namely *Enteromius cf. anoplus/motobensis* within the Potspruit river system on this farm. The presence of the alien Rainbow trout (*Oncorhynchus mykiss*) was also confirmed (visual observation) in the larger dam on the property. The barb (*E. cf. anoplus/motebensis*) requires further verification (Genus currently under review in RSA) and until verified the it will be considered to potentially be a fish species of conservation concern (due to E. motebensis (IUCN) listing as near-threatened (NT).

The current study also confirmed a high aquatic macroinvertebrate diversity present with the BDPNR.

At least one species of aquatic macroinvertebrate of conservation concern could be expected within the DBPNR study area, namely, *Pseudagrion newtoni* (VU: Vulnerable) *(Damselfly:* Harlequin sprite). The presence of this species was not confirmed during the current survey but there is a high probability that this species may be present within the DBPNR. The presence of various rare, endemic or range-restricted macroinvertebrate taxa were also confirmed. A total of forty-seven (47) macroinvertebrate families were also sampled within the DBPNR study area between 2020 and 2022. This reflects a relatively high diversity of aquatic macroinvertebrate families and is a reflection of highly diverse aquatic habitats as well as areas with very good water quality. Five taxa/groups with a *high* and eleven (11) taxa with a *moderate* requirement for unmodified water quality was present further confirming excellent water quality prevailing at present in most reaches of the DBPNR study area

Fifty-nine (59) diatom species were identified (first 400 counted per sample) at the five sampling sites assessed in DBPNR during February 2022. Four of the five sites were characterised by **high biological water quality** reflecting near pristine conditions, while the remaining site was rated as having moderate biological water quality. Endemic species with a preference for high biological water quality were observed. These species, based on the experience of the diatomologist, are scarce and have only been observed in the upper reaches of high altitude streams or the upper reaches near the origins of streams where anthropogenic activity is limited.

The current study highlighted a few potential impacts, threats or risks to the aquatic biodiversity diversity of the DBPNR . These potential impacts were described, an indication provided of the relevance to specific BMU's as well as the species or aspects of concern regarding the specific impact or threat.

Recommendations are made regarding potential management and mitigations measures that can be considered for implementation in an attempt to address and prevent any future impacts on the aquatic biodiversity of the DBPNR.

According to Hermoso *et al. (*2016), declaring protected areas (PAs) (such as the proposed De Berg Private Nature Reserve) stands out as one of the main conservation strategies worldwide and there are clear commitments to expand their extent under the auspices of the Convention on Biological Diversity. This conservation strategy has also received increasing attention in a freshwater context in the last two decades. Despite increasing conservation efforts, the effectiveness of PAs for freshwater purposes is questioned and freshwater biodiversity continues to decline. There are many reasons for this poor effectiveness: a lack of consideration of freshwater needs when designing and declaring protected areas, fewer resources devoted to freshwater conservation management than to other actions, and poor understanding of complex management problems beyond the limits of the protected area. Hermoso *et. al* (2016) advocate better monitoring programmes to assess the effectiveness of PAs for freshwater biodiversity, in which the unique characteristics of freshwater systems, such as the important role of connectivity and the close links with the rest of the landscape they drain, are considered. There are new conservation opportunities to enhance the value of PAs for freshwater biodiversity under the new conservation paradigm of 'people and nature'. The imperative of finding solutions that generate co-benefits alongside biodiversity conservation, and the clear reliance of human communities on freshwater services, has created an environment that may be more favourable to PAs focused in whole or part on fresh waters. The DBPNR as a proposed future protected area can therefore play an integral part in freshwater biodiversity conservation on a local, provincial and national scale.

Recommended future studies:

1. Focussed aquatic studies and biomonitoring in the De Berg Private Nature Reserve (DBPNR).

Due to the importance of the DBPNR in terms of aquatic biodiversity conservation and its value to conserve these upper catchments (especially the Groot Dwars River) to ensure continued good water quality and flow, it is recommended that aquatic assessments should be continued in this area. The following should be considered:

- Implement a biomonitoring programme (fish, SASS5, diatoms) at selected sites within the DBPNR to expand the spatial and temporal information regarding the aquatic biodiversity of this area and to monitor any potential impacts.
- Conduct more detailed fish assessments of the Potspruit and dams on the farm Goedehoop to verify the fish species composition of this area.

2. **Further studies on *E. cf. motobensis/anoplus***:

The barb species that closely resembles *Enteromius motebensis* (the Marico barb) and *E. anoplus* (Chubbyhead barb) was sampled from the Potspruit on the farm Goedehoop (BMU 5 and 7). This *Enteromius* species is potentially a unique genetic linage of the complex "*Enteromius anoplus/motebensis group of species"*. Previous genetic (unpublished) studies of the *Enteromius* species within the Northam Booysendal study area (Groot Dwars River) suggest that this population is genetically unique, as a result of its isolated distribution (Dr. Francois Roux, MTPA, *pers. comm.).* The taxonomy of this *Enteromius* species in the DBPNR remains uncertain and should as a matter of urgency be addressed by further studies. These studies should aim to verify the taxonomy of this species (including genetic analyses), investigate the current range of distribution and relative abundance within the study area, describe its preferred habitat and water quality and also identify specific threats and impacts. All this information should then be used to compile a detailed management plan if this species is confirmed to be of conservation importance.

9. REFERENCES

BARBOUR MT and STRIBLING JB (1994) A technique for assessing stream habitat structure. Pp. 156-178, In: Proceedings of the conference "Riparian Ecosystems of the Humid U.S. Management, functions, and Values". National Association of Conservation Districts. Washington, DC

CLEAN STREAM BIOLOGICAL SERVICES (CSBS) (2021). Northam Booysendal Platinum Aquatic Fauna Diversity, compiled as part of First Phase BMP study (2020/21). Report no CSBS/NB/2021/A.

DALLAS H. F. and DAY J (1993) The effect of water quality variables on Riverine Ecosystems: A Review. WRC Report Nr. TT 61/93.

DEPARTMENT OF WATER AFFAIRS AND FORESTRY (1996). South African Water Quality Guidelines (second edition). Volume 6: Agricultural water use: Aquaculture.

DEPARTMENT OF WATER AFFAIRS AND FORESTRY (1999). Resource Directed Measures for Protection of Water Resources. Volume 1. River Ecosystems Version 1.0, Pretoria

DICKENS C and GRAHAM M (2001) South African Scoring System (SASS) Version 5 Rapid Bioassessment Method for Rivers. River Health Programme Web Page.

HERMOSO V, ABELL R, LINKE S and BOON P (2016). The role of protected areas for freshwater biodiversity conservation: challenges and oppurtunities in a rapid changing world. *Aquatic Conserv: Mar. Freshw. Ecosyst. 26 (Suppl. 1): 3–11 (2016)*

KEMPSTER, P. L. HATTINGH, W. H. J. & VAN VLIET, H. R (1982). Summarised water quality criteria. Technical report NR. Tr 108. Department of Environmental Affairs.

KLEYNHANS, C. J. (1997). An exploratory investigation of the Instream Biological Integrity of the Crocodile River, Mpumalanga, as based on the Assessment of Fish Communities. Draft Report, Department of Water Affairs and Forestry, Institute for Water Quality Studies. 61 pp.

KLEYNHANS, C. J. (1997). An exploratory investigation of the Instream Biological Integrity of the Crocodile River, Mpumalanga, as based on the Assessment of Fish Communities. Draft Report, Department of Water Affairs and Forestry, Institute for Water Quality Studies. 61 pp.

KOEKEMOER AQUATIC SERVICES (KAS) (2022). De Berg Diatom Community Composition and biological water quality. Report by S. Koekemoer.

NEL J.L., A DRIVER, WF STRYDOM, A MAHERRY, C PETERSON, L HILL, DJ ROUX, S NIENABER, H VAN DEVENTER, E SWARTZ and LB SMITH-ADAO (2011). Atlas of Freshwater Ecosystem Priority Areas in South Africa: Maps to support sustainable development of water resources. WRC Report No. TT 500/11.

NEPID (2022). De Berg Private Nature Reserve: Preliminary assessment of aquatic macroinvertebrates. Report by R. Palmer.

RIVERS FOR AFRICA (RFA) (2011). Review and update of the Desktop Present Ecological Status (PES) and Ecological Importance (EI) and Ecological Sensitivity (ES) of South African Rivers according to the Sub-Quaternary (SQ) Catchments: OLIFANTS WMA. WRC PROJECT NUMBER: K5/2041.

SCHMIDT, RC and BART, HL (2015). Nomenclature changes should not be based on equivocally supported phylogenies: Reply to Yang *et al*. 2015. Molecular Phylogenetic and Evolution 90 (2015) 193-194.

SKELTON PH, (2001) A complete guide to freshwater fishes of Southern Africa. Struik Publishers (Pty) Ltd., Cape Town, South Africa. 395pp.

THIRION, C. A.; MOCKE, A & WOEST, R (1995). Biological Monitoring of Streams and Rivers using SASS4: A User Manual. Final Report, No. N 000/00/REQ/1195. Institute of Water Quality Studies, Department of Water Affairs and Forestry.

WOODFORD, D (2017). *Enteromius motebensis*. *The IUCN Red List of Threatened Species* 2017: e.T2581A100145924. https://dx.doi.org/10.2305/IUCN.UK.2017-3.RLTS.T2581A100145924.en. Downloaded on 14 June 2021.

YANG L, Sado T, Hirt MV, Pasco-Veil E, Arunachalam M, Li J, Wang X, Freyhof J, Saitoh K, Simins AM, Miya M, He S, Mayden RL (2015). Phylogeny and polypoidy: Resolving the classification of cyprinine fishes (Teleostei: Cypriniformes). Mol. Phylogenet. Evol. 85, 97 – 116.