

1 EcoTWIN 1.0: A Fully Distributed Tracer-Aided Ecohydrological Model Tracking Water, 2 Isotopes, and Nutrients

3 Songjun Wu¹, Doerthe Tetzlaff^{1,2}, Yi Zheng³, Chris Soulsby⁴

4 1 Department of Ecohydrology and Biogeochemistry, Leibniz Institute of Freshwater Ecology and
5 Inland Fisheries, Berlin, Germany

6 2 Geography Institute and IRI THESys, Humboldt University of Berlin, Berlin, Germany

7 3 School of Environmental Science and Engineering, Southern University of Science and Technology,
8 Shenzhen, China

9 4 Northern Rivers Institute, School of Geosciences, University of Aberdeen, UK

10 Corresponding author: Songjun Wu (songjun.wu@igb-berlin.de)

11

12 Abstract

13 The value of stable water isotopes in constraining process representation in hydrological models is
14 well acknowledged with numerous tracer-aided hydrological models developed in recent years, yet
15 few have leveraged these benefits for more robust water quality modelling. Therefore, we introduce
16 EcoTWIN, a fully distributed tracer-aided ecohydrological model that simultaneously **tracks water**,
17 **isotope**, and **nutrient** fluxes. A thorough model test was conducted by calibrating EcoTWIN against
18 discharge, in-stream isotopes, and NO₃-N concentrations (1980-2024) in 17 large-scale (10³ - 10⁵ km²)
19 European catchments spanning a wide range of geographic and climatic gradients. Furthermore, three
20 reanalysis products (ERA5 snow depth, MODIS evapotranspiration, and GRACE surface water anomaly)
21 were employed to further validate the capacity of EcoTWIN to reproduce associated but uncalibrated
22 internal water fluxes. Results showed good model performance of both calibrated in-stream targets
23 and uncalibrated internal fluxes in most catchments. Therefore, we conclude that EcoTWIN is a flexible,
24 transferable modelling tool for prediction and process inference in terrestrial ecosystems ranging from
25 boreal to subtropic climates. Constrained by tracer simulations, the model not only captures the
26 celerity, but also the velocity of hydrological fluxes, thus providing spatio-temporally-explicit
27 estimations of water ages and travel times. Such information provides opportunities to bridge
28 catchment hydrology and water quality by linking travel times with biogeochemical processing kinetics.
29 We demonstrate this with a proof of concept using Damköhler Number in nitrogen modelling.

30

31 1 Introduction

32 The development of ecohydrological models has been accelerating in the recent decades towards
33 frameworks that are more spatially distributed (instead of lumped or semi-distributed) and complex
34 (integrating more ecohydrological processes) (Pechlivanidis et al., 2011; Wellen et al., 2015). A few
35 examples include SWAT (Arnold et al., 2012), HYPE (Lindström et al., 2010), and mHM-Nitrate (Yang
36 et al., 2018), which have been widely applied worldwide. As process-based models, they are used not
37 only as prediction tools for specific variables, but also as learning tools for model inference, i.e., to

38 track the internal states/fluxes from available observations (Wang et al., 2024). This, however, poses
39 challenges due to the considerable uncertainties in model inference.

40 Inference of internal processes is naturally uncertain due to the lack of direct observations, though
41 such uncertainty can be constrained to some extent by rigorous split-sample calibration and validation.
42 The reason we use “somehow” here is based on the fact that most models are calibrated to a minimal
43 number of variables, and 81% of calibrations used data from a single gauge (mostly at a catchment
44 outlet) as reviewed in Wellen et al., (2015). Additionally, from a technical perspective, “equifinality”
45 further adds to the inference uncertainty due to the potential misinformation in data (uncertainty in
46 model forcing and observations) and model structure (the use of simplified, abstract mathematics to
47 simulate real world processes) (Beven, 2006). This can result in inaccurate process representations
48 yielding deceptively good results through error compensation, thus leading to overconfidence in a
49 model's ability to reproduce within-basin dynamics (Wen et al., 2024; Wu et al., 2025a). As
50 acknowledged by the hydrological community, models calibrated solely against discharge at the
51 catchment outlet reflect only the celerity of hydrological systems (pressure wave propagation), yet
52 constituent transport in water quality modelling relies on the velocity (mass flux of the water)
53 (McDonnell & Beven, 2014). Failure to reconcile these differences can lead to questionable process
54 inferences from many ecohydrological and water quality models.

55 One way to strengthen model inference is to include auxiliary data for calibration (Efstratiadis &
56 Koutsoyiannis, 2010). However, there is a paradox in multi-criteria calibration, as on the one hand,
57 more auxiliary data will feed unique information to the calibration process, thus effectively
58 constraining the model behaviour from an ecohydrological perspective; yet on the other hand, it
59 increases the dimensionality of calibration thus resulting in degraded performance or failure of
60 calibration from a technical perspective. The “curse” of dimensionality in ecohydrological modelling is
61 universal for all the commonly used algorithms under both Bayesian and Pareto theories as
62 demonstrated in Wu et al., (2025c). Therefore, modellers should expect the selected auxiliary data to
63 contain as much information as possible (Nearing et al., 2020). For distributed modelling, the auxiliary
64 data should reflect the cumulative contribution of all upstream reaches/regions, rather than variables
65 that are highly dependent on local condition/processes (e.g. point-scale soil moisture and
66 evapotranspiration measurements etc.).

67 Stable water isotopes, in this context, have powerful potential in cumulative flux tracking. As
68 conservative tracers, ^2H and ^{18}O are independent of biogeochemical reactions and naturally integrate
69 landscape heterogeneity, thus providing effective constraints on spatially distributed (dis)connections
70 of hydrological flow paths as well as velocity of the hydrological systems which reflect flux-storage
71 interactions (Jung et al., 2025; Tetzlaff et al., 2015). The value of tracers has long been recognised by
72 hydrologists (Hooper et al., 1988), with many tracer-aided hydrological models developed and evolved
73 in recent years from lumped (Birkel et al., 2011; Godsey et al., 2010), to semi-distributed (van
74 Huijgevoort et al., 2016; Nan et al., 2021), and distributed structure (Kuppel et al., 2018; Remondi et
75 al., 2018). However, few attempts have been made to integrate a tracer-aided hydrological structure
76 into water quality modelling (Birkel & Soulsby, 2015; Jung et al., 2025), despite the need being evident

77 for nearly four decades (Neal et al., 1988). Moreover, existing pioneering models are mostly
78 conceptualised/lumped (Benettin et al., 2015; Dick et al., 2015) and/or loosely coupled via external
79 tracer/water quality modules (Pesántez et al., 2023; Yang et al., 2024; Zhang et al., 2020). The external
80 coupling of model chains transfer necessary internal states and fluxes between sub-models (e.g.
81 hydrological fluxes for constituent mixing in water quality or isotopic modules) via online in-memory
82 coupling (instead of offline on-disk coupling), thus significantly increasing the resources consumption
83 in input/output operations. Such model chains, though providing useful scientific insights, can become
84 problematic for large-scale applications owing to the exponential growth in computational and
85 storage requirements. Therefore, there remains a need to develop a fully distributed, computationally
86 efficient ecohydrological model that combines hydrological, isotopic, and water quality simulations.

87 This research gap motivated the development of EcoTWIN, the model that we present in this paper.
88 To our knowledge, the model is one of the first distributed tracer-aided **ecohydrological** models that
89 **tracks water, isotopic, and nutrient** fluxes simultaneously in a C++-based framework. For a thorough
90 testing of EcoTWIN, 17 large European catchments were selected for calibration against discharge, in-
91 stream isotopes, and NO₃-N concentrations. These catchments span over a wide range of geographic
92 (Alpine to lowland plain) and climatic (from snow-dominated to Mediterranean) gradients. In addition,
93 the robustness of modelled inference on uncalibrated internal fluxes were also compared with three
94 remote sensing products (snow depth, evapotranspiration, and water storage). Given the overall good
95 integrated performance in most catchments, EcoTWIN is presented as an ecohydrological modelling
96 framework applicable for terrestrial ecosystems ranging from boreal to temperate and subtropical
97 climates across a wide range of geographical environments. The subsequent sections are organised as
98 follows: Section 2 and 3 introduce the model structure of EcoTWIN and details in calibration and
99 validation; the model performance is shown in Section 4; in Section 5 we show the advantages of a
100 tracer-aided ecohydrological framework with an example of how water ages bridge catchment
101 hydrology and water quality models; finally, the current limitations and planned future development
102 of EcoTWIN are also discussed.

103

104 **2 Model description**

105 EcoTWIN is fully distributed ecohydrological model implemented in C++. The model consists of
106 hydrological, isotopic, and nitrogen modules, which simulate major ecohydrological states and fluxes
107 from canopy to groundwater (Figure 1). Aided by tracer simulations, the model is additionally able to
108 track the water movement vertically and laterally via the calculation of water ages and travel times.
109 For detailed information of model parameters please refer to Table S1.

110

111 **2.1 Hydrological module**

112 EcoTWIN follows a typical multi-layer, top-down, bucket-type approach that resolves the water
113 balance sequentially for the vegetation canopy, three soil layers, unsaturated zone, and groundwater.

114 As the foundation of solute transport, the hydrological module employs a selective disassembly
 115 structure with multiple alternative conceptualisations possible for specific important hydrological
 116 processes. The configuration can be specified *a priori* based on the goal of modelling and prior
 117 knowledge of the studied catchment(s).

118 **2.1.1 Soil properties**

119 Before iterative simulations, soil characteristics are estimated using appropriate pedotransfer
 120 functions. Three different alternatives are provided, each of which requires different levels of inputs
 121 but all were found to provide robust estimation of soil porosity (θ_s), field capacity (θ_{fc}), wilting point
 122 (θ_{wp}), and hydraulic conductivity (K_s). All the soil properties are required for each soil layer/depth.
 123 This can be achieved via three alternative options: (i) assigning identical properties across the whole
 124 soil column, (ii) calculating separately for each depth based on depth-dependent inputs, or (iii)
 125 extrapolating deeper profile characteristics from the top soil properties based on a depth-dependent
 126 equation in Maneta & Silverman, (2013).

127 The distribution of soil types and land use is assigned from raster file in EcoTWIN. This can be specified
 128 as a static boundary condition; alternatively, the distributions can also be updated dynamically via a
 129 user-specified interval to reflect any temporal changes due to land management.

130

131 **2.1.2 Vertical fluxes**

132 The vertical fluxes are resolved for storages in the canopy, soil layers, unsaturated zone, and
 133 groundwater. The mass balance of canopy storage (ΔC) follows:

$$\Delta C = P - I - Th \quad (1)$$

134 where P , I , Th are precipitation, interception, and throughfall, respectively. The throughfall is
 135 calculated as the exceedance of current canopy storage from the maximum storage calculated by Leaf
 136 Area index LAI and a correlation parameter α .

$$C_{max} = \alpha * LAI \quad (2-1)$$

137 Alternatively, the maximum canopy storage can be estimated with explicit consideration of
 138 precipitation intensity (Landgraf et al., 2023):

$$C_{max} = \alpha * LAI * \left(1 - \frac{1}{1 + SCF * P / (\alpha * LAI)}\right) \quad (2-2)$$

139 where SCF is the vegetation cover fraction calculated by LAI and an extinction coefficient (rE)
 140 adopted from HYDRUS-1D (Šimůnek et al., 2013):

$$SCF = 1 - \exp(-rE * LAI) \quad (3)$$

141 Then throughfall is calculated as the exceedance of canopy storage from the maximum:

$$Th = (P + C) - C_{max} \text{ if } (P + C) > C_{max} \text{ else } 0 \quad (4)$$

142 After reaching land surface, throughfall becomes ponding water (S_{Pond}) or snow (S_{Snow}) depending
 143 on a temperature threshold for separation ($Thre_{SN}$). Snow will melt and recharge the ponding water
 144 in warm conditions (air temperature Ta exceed $Thre_{sn}$) following a degree-day model.

$$melt = S_{snow} * \min(dd_{min} + dd_{inc} * Th * (Ta - Thre_{SN}), dd_{max}) \quad (5)$$

145 Where dd_{min} and dd_{max} are the minimum and maximum of degree day factor, while dd_{inc} denotes
 146 the rate of increase in the degree-day factor per degree Celsius rise in temperature.

147 The available ponding water infiltrates into the top soil layer using Green-Ampt model (Kale & Sahoo,
 148 2011; Maneta & Silverman, 2013), with infiltration capacity first calculated as a function of average
 149 soil moisture over the hydrologically active depth:

$$I_f = Ks * \left(1 + \frac{\psi * \theta_s * (1 - (\theta_1 - \theta_{wt}) / (\theta_s - \theta_{wt}))}{\theta_1 * d_1}\right) \quad (6)$$

150 Where θ_1 , θ_s , θ_{wt} , and d_1 are the moisture content, porosity, wilting point, and depth in top soil layer;
 151 ψ is a parameter representing soil air entry pressure in m. Then potential infiltration (F_p) is determined
 152 from the lesser between the available ponding water (S_{pd}) and potential infiltration rate integrated
 153 over time before ponding occurs ($I_f * t_p$).

154 The actual infiltration (F) is solved iteratively using the Newton–Raphson scheme:

$$F = \Delta\theta * d_1 = F_p + Ks * w_{Ks} * (\Delta t - t_p) - \psi\Delta\theta * \ln\left(\frac{\psi\Delta\theta + \Delta\theta d_1}{\psi\Delta\theta + F_p}\right) \quad (7)$$

155 where w_{Ks} is anisotropy ratio of vertical to horizontal Ks .

156 The soil storage in each layer is conceptualised as two water pools – a gravitational, free-flowing pool
 157 and a capillary, soil-bound pool. The two pools are separated based on field capacity (Maneta &
 158 Silverman, 2013), and percolation happens when soil storage exceeds the threshold. Three alternative
 159 schemes are included in EcoTWIN.

160 In the first scheme, all water in excess of field capacity percolates to deeper layer:

$$Pc_i = (\theta_i - \theta_{fc}) * d_i \quad (8-1)$$

161 where Pc_i , θ_i and d_i depict the percolation, moisture content and depth from/in i th soil layer in m.

162 The second scheme additionally considers the hydraulic conductivity (Ks) following the
 163 conceptualisation in SWAT (Arnold et al., 2012):

$$Pc_i = (\theta_i - \theta_{fc,i}) * d_i * \left(1 - \exp\left(\frac{-\Delta t * Ks}{\theta_{s,i} - \theta_{fc,i}}\right)\right) \quad (8-2)$$

164 The third scheme relates percolation to the extent of soil saturation with an exponential parameter β
 165 (Kumar et al., 2013; Samaniego et al., 2010):

$$Pc_i = (\theta_i - \theta_{fc,i}) * d_i * \left(1 - \exp(\beta * \log(\theta_i / \theta_{s,i}))\right) \quad (8-3)$$

166 For evapotranspiration, soil evaporation and transpiration are estimated separately. The separation
 167 of PET is realised by surface cover fraction introduced above:

$$PT = PET * SCF; \quad PE = PET - PT \quad (9)$$

168 Soil evaporation is simulated in the top soil layer based on the soil saturation:

$$Evap_s = PE * \min\left(\frac{\theta_1}{\theta_{fc,1}}, 1\right) \quad (10)$$

169 Transpiration is simulated in all soil layers based on the fractions ($f_{root,i}$) of root density ($D_{root,i}$) in
 170 each layer partitioned by soil depth and a parameter (γ_{root}):

$$Tr_i = PT * f_{root,i} * \frac{\theta_1 - \theta_{wp,1}}{\theta_{fc,1} - \theta_{wp,1}} \quad (11)$$

$$f_{root,i} = D_{root,i} / \sum_{j=1}^m D_{root,j} \quad (12)$$

$$D_{root,i} = \left(1 - \gamma_{root} \left(\sum_{j=1}^m d_j\right)\right) - \left(1 - \gamma_{root} \left(\sum_{j=1}^i d_j\right)\right) \quad (13)$$

171 Channel evaporation is also estimated using Penman equation, which relies on net radiation, wind
 172 speed, air pressure, and air temperature as inputs.

173 The last soil layer percolates to an unsaturated storage in unsaturated zone (S_{unsat}). The
 174 compartment stores the excess water from soil and percolates either downward to groundwater
 175 storage (S_{GW}) or laterally downstream. The percolation to groundwater P_{cGW} is determined by a
 176 weighting parameter p_{GW} as a proportion of unsaturated storage:

$$P_{cGW} = S_{unsat} * p_{GW} \quad (14)$$

177 Additionally, irrigation is conceptualised in EcoTWIN, which is realised via the water extraction from
 178 river or groundwater. The source is determined by the geographic location: for a grid cell with channel
 179 network, water is extracted directly from river, and local groundwater is used as irrigation source for
 180 non-channel grids. The amount of extraction is estimated from a predefined coefficient for crop water
 181 demands (w_{irr}) from which the deficit is calculated for each of the m soil layers.

$$deficit = \sum_{i=1}^m (\theta_{fc,i} - \theta_{wp,i}) * w_{irr} * d_i \quad (15)$$

182 Note that the irrigation can switch to groundwater extraction if river storage cannot fill the deficit.

183

184 **2.1.3 Lateral fluxes**

185 In EcoTWIN, grid cells are connected laterally at three levels - surface, unsaturated zone, and
 186 groundwater. Note that some models omit the unsaturated storage and directly calculate excess
 187 water to drain based on the saturation extent of the bottom soil layer (e.g., ECH₂O-iso, Kuppel et al.,

188 2018). EcoTWIN did not follow this conceptualisation because in reality, the lateral drainage is focused
 189 in the saturated zone, and thus the bottom of the soil layer instead of the whole soil profile. The
 190 drainage of an entire soil layer thus brings considerable uncertainty to the velocity of lateral transport
 191 when the lower boundary of the soil is a parameter to tune in calibration. For instance, a large soil
 192 depth will dramatically reduce the velocity of interflow drainage and slow down the mixing of
 193 constituents, though this might still perfectly reproduce the celerity (hydrograph) for purely
 194 hydrological modelling. Our conceptualisation (an independent unsaturated compartment) aligns
 195 with most hydrological models (Arnold et al., 2012; Yang et al., 2018) and fits the recent analysis
 196 supporting the dominant role of lateral drainage over vertical transports globally (Mcmillan et al.,
 197 2025).

198 By the end of each timestep, ponding water receives upstream inputs and contributes to channel
 199 storage if the grid is connected to the channel network, while non-channel grid has $Ovf_C = 0$:

$$Ovf_C = (Ovf_{T,in} + S_{pond}) * p_{Ovf} * dx_C/dx_T \quad (16)$$

200 dx_C and dx_T are the channel length and size of terrestrial grid cell; p_{Ovf} is a weighting parameter for
 201 channel recharge. Then the remaining ponding water routes to downslope terrestrial grid following
 202 the topographic gradient. In none-channel grid cells, all available ponding storage routes lateral
 203 downstream ($Ovf_C = 0$):

$$Ovf_{T,out} = (Ovf_{T,in} + S_{pond}) - Ovf_C \quad (17)$$

204 Similarly, unsaturated storage contributes first to channel storage in grid cells within channel network,
 205 while non-channel grid cells have $Inf_C = 0$:

$$Inf_C = (Inf_{T,in} + S_{vadose}) * K_{vadose} * \left(1 - e^{-1 * exp_{Inf} * (Inf_{T,in} + S_{vadose})}\right) * p_{Inf} \quad (18)$$

206 where K_{vadose} is the effective conductivity of lateral transport in the unsaturated zone; while exp_{Inf}
 207 is an exponential parameter determining the strength of positive correlation between recharge and
 208 current unsaturated storage. Then the remaining unsaturated storage is partially routed to downslope
 209 grid cell following a linear approximation of Kinematic wave equation, which assumes gravitational
 210 flux per unit width $Inf_{T,out}$ is proportional to the subsurface hydraulic conductivity (K_{vadose}) and
 211 bedrock slope ($slope$ approximated from the surface slope):

$$Inf_{T,out} = (Inf_{T,in} + S_{unsat} - Inf_C) * \left(1 + \alpha * \frac{dt}{dx}\right) * \alpha * \frac{dt}{dx} \quad (19)$$

$$\text{where } \alpha = K_{unsat} * \sin(slope)$$

212 Groundwater routing is similar to that of interflow, with channel recharge followed by terrestrial
 213 transport. Note that the terrestrial groundwater flow does not consider the bedrock slope as
 214 groundwater storage is generally much large than unsaturated storage, and thus independent from
 215 topographic gradients:

$$GWf_C = (GWf_{T,in} + S_{GW}) * K_{GW} * \left(1 - e^{-1 * exp_{GWf} * (GWf_{T,in} + S_{GW})}\right) * p_{GWf} \quad (20)$$

$$GWf_{T,out} = (GWf_{T,in} + S_{GW} - GWf_C) * \left(1 + \alpha * \frac{dt}{dx}\right) * \alpha * \frac{dt}{dx} \quad (21)$$

where $\alpha = K_{vadose}$

216 The channel routing is realised using Kinematic wave equation based on a scaled channel roughness
217 parameter (Maneta & Silverman, 2013).

218

219 **2.2 Isotopic module**

220 The isotopic module in EcoTWIN tracks the composition of stable water isotopes in all water storage
221 compartments following hydrological mixing and fractionation. The module also provides estimation
222 of water age and travel time conceptualised as the time since water molecules enter the catchment
223 as precipitation, and the time water molecules need to travel through the specific storage.

224 **2.2.1 Mixing**

225 The mixing and transport of isotopes (^2H and ^{18}O , both noted as C) are governed by the velocity of
226 hydrological fluxes with a complete mixing strategy for most water storages:

$$\frac{d(V * C)}{dt} = \sum_{k=1}^{N_{in}} q_{in,k} * C_{in,k} - \sum_{k=1}^{N_{out}} q_{out,k} * C \quad (22)$$

227 Where V and C are the volume and composition/concentration of the storage, while N_{in} and N_{out}
228 denote the number of influx and outflux. Such strategy is built on two assumptions: (i) constitutes (i.e.,
229 isotopes) are fully mixing within each timestep; (ii) the composition/concentration in outflow equals
230 to that in storage. Additional mixing between ponding and upper soil water storage is allowed (with
231 proportion determined by a parameter *SrfMixing*), as nutrients in top soils can be flushed out in
232 large hydrological events (Seybold et al., 2022).

233 The full-mixing assumptions have been widely used and shown to be reasonable for storages with
234 relatively small volumes in many mixing/water quality models (Arnold et al., 2012; Yang et al., 2018).
235 However, some studies show that a complete mixing strategy can be problematic for large storages
236 such as groundwater as they are generally poorly constrained (e.g. Soulsby et al., 2015). Therefore,
237 the mass conservation equation used in the INCA-N model and mHM-Nitrate is employed to calculate
238 the mixing of groundwater storages with influxes (i.e., percolation from unsaturated storage and
239 lateral groundwater inflow).

$$\frac{dC}{dt} = \frac{1}{V + V_r} * \left(\sum_{k=1}^{N_{in}} q_{in,k} * C_{in,k} - \sum_{k=1}^{N_{out}} q_{out,k} * C \right) \quad (23)$$

240 where V_r is the retention storage. The equation is solved by the fourth order Runge-Kutta technique.

241

242 **2.2.2 Fractionation**

243 As conservative tracers, the composition of isotopes in water storages/fluxes is only changed by
 244 kinetic fractionation apart from hydrological mixing. The process is accompanied by evaporation,
 245 resulting in the preferential loss of lighter isotopes (^1H and ^{16}O) to the vapor phase and a
 246 corresponding enrichment of heavier isotopes (^{18}O and ^2H) in the residual water. In EcoTWIN, the
 247 fractionation is simulated along with evaporation of top soil water and river storage based on the
 248 Craig-Gordon model (Craig et al., 1964; Kuppel et al., 2018), while transpiration is assumed to be a
 249 non-fractionating process (Dawson & Ehleringer, 1991; Kuppel et al., 2018).

$$C = C^* - (C^* - C) * \left(\frac{S - Evap}{S} \right)^m \quad (24)$$

250 where C^* and m are the limiting isotopic composition (in ‰) and the dimensionless enrichment slope
 251 that are estimated via the following equations in (Good et al., 2014):

$$C^* = \frac{h_a C_a + h_s \varepsilon^+ + \varepsilon^k}{h_s - h_a + \varepsilon^k / 1000} \quad (25)$$

$$m = \frac{h_a - (h_s \varepsilon^+ + \varepsilon^k) / 1000}{h_s - h_a + \varepsilon^k / 1000} \quad (26)$$

252 where h_a is the relative humidity above the soil surface normalised from atmospheric relative
 253 humidity (h), air temperature (T_a), and soil temperature (T_s estimated from Amato & Giménez, 2024).
 254 C_a is the isotopic composition of ambient air moisture estimated from precipitation composition:

$$C_a = (C_{rain} - \varepsilon^+) / \alpha^+ \quad (27)$$

255 where ε^+ is the equilibrium fractionation factor (Skrzypek et al., 2015); α^+ is a temperature factor
 256 estimated from T_a .

$$\varepsilon^+ = (1 - 1/\alpha^+) * 1000 \quad (28)$$

257 The factor of diffusion-controlled kinetic isotopic separation ε^k is calculated based on the relative
 258 humidity of soil surface (h_a) and soil pore (h_s).

$$\varepsilon^k = (h_s - h_a) * \left(1 - \frac{D_i}{D} \right) * n \quad (29)$$

259 Where D_i and D denote the diffusivities of water vapor molecules containing heavier isotope and the
 260 lighter isotope, respectively. The ratio can be acquired in Horita et al., (2008) for ^2H (0.9877) and ^{18}O
 261 (0.9859). n is an advection term ranging between 0.5 (in saturated soils) and 1 (in dry soils). The factor
 262 is included in calibration for the fractionation of top soil evaporation yet fixed as 0.5 for that of channel
 263 evaporation.

264

265 **2.2.3 Water age and travel time**

266 EcoTWIN can track the age of water i.e., the time since water enters the catchment as precipitation,
267 in each storage. In age tracking, precipitation is defined as new water with age of zero. At the end of
268 each time step, water ages of all storages are advanced based on the temporal resolution (for instance
269 one day if the model is set up for daily timesteps). Note that in some circumstances, the modellers
270 might need to disable the age evolution of specific storage(s) (e.g., groundwater storage) as the
271 storage can be too large to achieve steady states in model spin-up. Similar to isotopes, water ages are
272 only controlled by hydrological transport with the same mixing strategy (i.e., complete mixing except
273 for groundwater).

274 The water ages in EcoTWIN are the mean values averaged from all water molecules in the storage,
275 which might be dominated by the inflow of very old water that obscure the age distribution of the
276 young water (e.g., the groundwater input to top soils due to the groundwater extraction for irrigation).
277 Therefore, EcoTWIN additionally provides the estimation of travel time - the time of water molecule
278 travelling through each storage. The simulation is similar to that of water ages. The only difference is
279 that the transition of water between storages (e.g., percolation into deeper soil layers) resets the travel
280 time to zero. Accordingly, all the water enters a new storage becomes new water instead of just
281 precipitation in water age tracking.

282

283 **2.3 Nitrogen module**

284 The nitrogen module describes the mass balance of nitrogen, particularly nitrate as the main form of
285 dissolved nitrogen, which is dominated by the interaction of hydrological transport and
286 biogeochemical transformations.

287 For each timestep, the nitrate concentration is simulated in each storage following three processes –
288 hydrological transport/mixing, nitrogen inputs, and biogeochemical transformations. Fully integrated
289 with hydrological module, nitrate transport also aligns with hydrological fluxes following the same
290 mixing strategy as in the isotopic simulation. For nitrogen sources, EcoTWIN considers the inputs from
291 fertiliser, manure, and plant residues, whose annual inputs can be specified via configuration. Notably,
292 fertilization can be parameterised via spatial raster inputs if corresponding dataset is available. The
293 timing and extent of nitrogen addition of all sources are determined following the implementation in
294 HYPE (Lindström et al., 2010), which distributes the annual sum across a specified period (e.g., the
295 period between planting and harvest for crops). Additionally, wet deposition is conceptualised as the
296 atmospheric nitrogen source, whose concentration can be specified via spatial raster and simply as a
297 constant value.

298 The biogeochemical transformations are mainly modified from the mHM-Nitrate model (Yang et al.,
299 2018), and the HYPE model (Lindström et al., 2010), which are conceptualised for the soil profile and
300 channel network. In the soil profile, three nitrogen pools are conceptualised for each soil layer,
301 including an inactive nitrogen pool (SN_i), an active nitrogen pool (SN_a), and a dissolved nitrate pool

302 (DN). The soil transformations include degradation (Dgd_s , from SN_i to SN_a), mineralisation ($Minr_s$,
 303 from SN_a to DN), denitrification ($Deni_s$, from DN to gaseous N_2), and plant uptake ($Uptk_s$, DN
 304 removal).

$$Dgd_s = SN_i * ref_{Dgd,s} * f_{Ta} * f_{\theta} / dt \quad (30)$$

$$Minr_s = SN_a * ref_{Minr,s} * f_{Ta} * f_{\theta} / dt \quad (31)$$

$$Deni_s = DN * ref_{Deni,s} * f_{Ta} * f_{\theta,deni} * f_{conc,s} / dt \quad (32)$$

305 where $ref_{Dgd,s}$, $ref_{Minr,s}$, $ref_{Deni,s}$ are the parameters representing the reference rates of soil
 306 degradation, mineralisation, and denitrification. f_{Ta} and f_{θ} are the regulating factors of temperature
 307 and moisture.

$$f_{Ta} = 2^{(T_a - 20)/10} * \omega \quad \text{where } \omega = \begin{cases} 1 & T_a > 5 \\ T_a/5 & 0 \leq T_a \leq 5 \\ 0 & T_a < 0 \end{cases} \quad (33)$$

$$f_{\theta} = \min \left[\frac{(1 - p_{\theta,deni}) * (\theta_{fc,i} - \theta_i)}{p_{\theta,fc} * d_i}, \frac{(\theta_i - \theta_{wp,i})}{p_{\theta,wp} * d_i} \right] \quad (34)$$

308 where $p_{\theta,fc}$ and $p_{\theta,wp}$ are the empirical factors that are fixed as 1.2, 0.8 based on literature values
 309 (Lindström et al., 2010; Yang et al., 2018). $p_{\theta,deni}$ is the saturation threshold for soil denitrification
 310 ranging between 0.4 – 0.85 (Yang et al., 2018). A different moisture factor considering a saturation
 311 threshold (θ_{thres}) is employed for denitrification, as denitrification is more sensitive to the soil
 312 wetness condition:

$$f_{\theta,deni} = [(\theta_i / \theta_{fc,i} - \theta_{thres}) / (1 - \theta_{thres})]^{2.5} \quad (35)$$

313 The process is additionally controlled by the concentration level in the storage $f_{conc,s} = C / (C + 10)$.
 314 Plant uptake is simulated using a three-parameter logistic growth equation in (Eckersten et al., 1994;
 315 Lindström et al., 2010).

316 Currently, in-stream denitrification is the only process considered in EcoTWIN.

$$Deni_w = ref_{Deni,w} * f_{Tw} * f_{conc,w} * A / dt \quad (36)$$

317 where $ref_{Deni,w}$ is the reference in-stream denitrification rates. The actual rates are regulated by a
 318 concentration factor $f_{conc,w} = C / (C + 1.5)$ and a temperature factor f_{Tw} (the same equation for f_{Ta}
 319 with inputs substituted by river temperature f_{Tw} , simplified as the rolling-average of 20-day air
 320 temperature).

321 It should be noted that the calibrated soil depth in this study is about 2.5 m, with intermittent
 322 saturation occurring in the deeper layer. This means that terrestrial denitrification is a combination of
 323 soil and groundwater processes in this study, though this might change in other applications if a
 324 shallow soil depth is assigned.

325

326 **3 Model calibration and validation**

327 A robust model application should not only reproduce observed variables through calibration but also
328 yield realistic estimates of internal states and fluxes that are not included in the calibration process.
329 This is essential to avoid situations where inaccurate process representations produce deceptively
330 good results through error compensation. Therefore, we evaluate EcoTWIN from both perspectives.
331 First, we assess the model's ability to reproduce observations via calibration (methods and results in
332 Sections 3.2 and 3.4). Then, we examine the model's capacity to simulate uncalibrated internal states
333 and fluxes by comparing the simulated snow depth, evapotranspiration, and total water storage with
334 corresponding remote-sensing products (methods and results in Sections 3.3 and 3.5).

335 To ensure model generality, 17 catchments were selected for calibration and validation depending on
336 the data availability (particularly stream stable water isotopes and nitrate), which span a wide range
337 of characteristics in geography, climate, and anthropogenic managements (Figure 2 and Table 1).
338 Anthropogenic management practices have a less dramatic effect than climate and geography in most
339 catchments due to the relatively low proportion of urbanized areas. However, a few notable
340 exceptions—such as the Rhine, Elbe, and Danube catchments—are included in the analysis, as these
341 densely populated regions hold critical ecological, agricultural, and economic importance for Europe,
342 and are subject to intensive human interventions in water management. This also provides a chance
343 to examine the applicability of EcoTWIN in human-affected catchments.

344

345 **3.1 Model setup**

346 EcoTWIN was setup for each of the 17 catchments for calibration with a spatial resolution of 5 km²
347 and a temporal resolution of daily timesteps from 1980 to 2024 (with first two years for spin-up). As
348 a fully distributed model, gridded GIS inputs are used in the model setup, including a digital elevation
349 model, flow direction, slope, channel width, channel length, proportion of each land use type (Winkler
350 et al., 2021), proportions of each soil type (world soil map, WRB2014), and soil properties (e.g., depth-
351 dependent proportions of clay, sand, silt, and organic matter from SOILGRIDS). All spatial inputs were
352 acquired with finer resolution (50 m or above) and resampled to the resolution of this application (5
353 km).

354 The climatic variables used to drive EcoTWIN include precipitation, air temperature, potential
355 evapotranspiration, relative humidity, and a few variables that are optional required for the
356 calculation of channel evaporation (air pressure, net radiation, and wind speed). These climatic
357 variables are available from the reanalysis products ERA5 and E-OBS, while PET is calculated using FAO
358 Penman-Monteith equation. For nitrogen simulations, additional inputs are needed including the
359 fertilization map (Grizzetti et al., 2021) and nitrate concentration of rainfall (Zhu et al., 2025) as the
360 boundary of nitrogen addition from agricultural activities and wet deposition.

361

362 3.2 Model calibration

363 **Method.** The calibration was conducted separately for each catchment to test the applicability of
364 EcoTWIN under different geological and climatic contexts. Three commonly used variables for
365 hydrological and water quality modelling (discharge, stream water isotope composition, and in-stream
366 NO₃-N concentrations) are employed for calibration. Their long-term time series were acquired at daily
367 steps from different sources (discharge from GRDC, isotopes from Wateriso and GNIR, and NO₃-N
368 concentration from global water quality database, GEMStat), and then compared with simulation
369 results at multiple sites for each catchment. Here ¹⁸O was selected for isotopic validation due to its
370 higher precision and data abundance. Given the discrepancy in duration of observations (especially
371 for isotopes and NO₃-N), a separate calibration and validation based on a split-sample approach is
372 difficult. Therefore, the full timescale (1982 - 2024) was used for calibration (and the validation
373 introduced in Section 3.3).

374 The DiffeREntial Evolution Adaptive Metropolis algorithm (DREAM) was selected for parameter
375 optimisation due to its relatively efficient and effective performance for high-dimensional problems
376 (as benchmarked in Wu et al., 2025c). The algorithm was implemented separately for each catchment
377 with the same prior distribution of parameters (Table S1). The maximum iteration was set as 100,000
378 for each catchment (20 chains with maximum chain length of 5000), from which 40 best simulations
379 were selected from the posterior distribution. The Kling-Gupta efficiency (KGE) statistic was used to
380 construct an informal likelihood function for DREAM optimisation.

$$l = \left[\sum_{i=1}^{N_{obs}} \sum_{j=1}^{N_{site}} (1 - KGE) * w_{i,j} \right]^{-m} \quad (37)$$

381 Where l is the likelihood; N_{obs} and N_{site} are the number of observation types (discharge, isotopes,
382 and nitrate) and sites. The weight $w_{i,j}$, defined for observation type i at site j , is assigned equally
383 across sites such that the total weight for each observation type sums to 1/3. m is an exponentially
384 coefficient to stretch the likelihood surface that is often set based on the number of observation points.
385 After prior test run, m was set as 500. Finally, the likelihood function is transformed to logarithmic
386 form for numeric stability. The calibration was validated using Kling-Gupta efficiency (KGE), Root Mean
387 Square Error (RMSE), Pearson Correlation Coefficient (Coefficient), and Percent bias (Pbias) (Table 2).

388 **Calibration performance.** EcoTWIN successfully reproduced the observed discharge in all 17
389 catchments with KGE exceeding 0.5 at most site (Figure 3a). Such performance is comparable to or
390 better than previous continental calibration of hydrological models (e.g., ParFlow, Naz et al., 2023; E-
391 HYPE, Donnelly et al., 2016).

392 Similarly, isotopic and nitrate simulations also produced good performances at most sites (Figure 3b).
393 However, there are a few exceptions. The failure of isotopic simulations was found at two sites within
394 the Alpine region (bottom left corner of figure 3b). This can be attributed to the uncertainty in
395 precipitation isotopes and snowmelt isotopes (due to the lack of snow elusion fractionation; Ala-aho
396 et al., 2017), the incorrect isotopic composition in groundwater, or the reduced applicability of degree-

397 day model for mountainous areas in Europe. Such simulation deviation due to the uncertainty in data
398 and boundary initialisation is often reported in previous calibration (Smith et al., 2021).

399 In general, the model produces comparable performances to existing nitrogen modelling at catchment
400 (Wu et al., 2022, 2025b; Yang et al., 2018) and continental scales (Jones et al., 2023; Mikayilov et al.,
401 2015). However, nitrate simulations failed to capture the observations at three sites, though as is
402 shown in Figure S1, these all have relatively low levels of NO₃-N concentrations. Such low average
403 values can easily trigger the degradation in KGE as one of the sub-components of KGE is highly
404 sensitive to the mean deviation, though the absolute deviation remained low (Figure S1 and Table 2).
405 Overall, we concluded that EcoTWIN has good capacity to reproduce in-stream components for a wide
406 range of catchments and for relatively long periods.

407

408 **3.3 Model validation**

409 **Method.** Remote sensing or reanalysis products were further employed to validate uncalibrated
410 internal model states or fluxes from three important perspectives in ecohydrological modelling – snow
411 depth from ERA5, evapotranspiration from MODIS, and surface water mass anomaly from GRACE (as
412 a storage proxy). The simulated variables corresponding to these products are, respectively, the depth
413 of snow pack, the sum of soil evaporation, channel evaporation, and transpiration from all soil layers,
414 and the anomaly of total water storage above groundwater (i.e., the sum of canopy storage, snow,
415 soil water storages, and unsaturated storage). The validation was realised via resampling the remote
416 sensing products to 5 km and comparing grid-to-grid with the modelled outputs. r^2 was used as the
417 performance metrics, as KGE is not applicable for time series with zero average, yet the average of
418 surface mass anomaly is close to 0.

419 Note that all three products may contain considerable uncertainties. ERA5 is a reanalysis product that
420 combines historical observations into global estimates using modelling and data assimilation
421 approaches, therefore inevitably embeds uncertainties associated with model structure and
422 observational coverage (Hersbach et al., 2020). MODIS evapotranspiration is derived from remotely
423 sensed spectral information, energy partitioning approaches and the Penman–Monteith framework,
424 whose uncertainty may exceed 30% depending on spatial scale and environmental conditions (Mu et
425 al., 2011). GRACE infers changes in terrestrially stored water masses from spatial and temporal
426 variations in the Earth’s gravity field; however, its coarse spatial resolution can introduce substantial
427 uncertainty when used for hydrological validation, particularly at basin or sub-basin scales (Tapley et
428 al., 2004). Nevertheless, good agreement between simulations and remote sensing or reanalysis
429 products can enhance confidence in the robustness of simulated spatial and temporal patterns,
430 although it does not necessarily imply accurate representation of absolute magnitudes.

431 **Validation performance.** First, we compared the sum of soil evaporation, channel evaporation, and
432 transpiration to MODIS evapotranspiration in each grid cell. The results in Figure 4 shows a general
433 good fit between simulation and MODIS records with KGE and r^2 above 0.5 in most regions. From the

434 subplots in Figure 4, we can see that the seasonality and magnitude of evapotranspiration were well
435 captured though the peaks in summer were slightly underestimated.

436 Then, the water storage anomaly was compared to the anomaly of simulated surface storage, i.e., the
437 sum of canopy storage, snow, soil water storages, and unsaturated storage. The grid-to-grid
438 comparison in Figure 4 shows a general good fit in most regions with r^2 close to or above 0.5. However,
439 more degradation was found compared to the performance in evapotranspiration, especially in
440 coastal regions. For instance, GRACE exhibited considerably increasing trends in water storage
441 between 2005 to 2015 in two Nordic catchments (W2 and W3), yet our simulations only showed a
442 moderate increasing trend. Similar degraded performance was found in the coastal catchments (e.g.,
443 three British catchments W6-8 in UK), though the magnitudes of simulation and GRACE data fit well.
444 This is possibly attributed to the coarse resolution of GRACE which additionally considered the storage
445 mass from ocean in coastal region yet not included in this terrestrial-explicit modelling.

446 Finally, the simulated snow depth was compared to the daily snow depth in ERA5 reanalysis products
447 (ERA5 post-processed daily statistics on single levels; 10.24381/cds.4991cf48). Results in Figure 4
448 show a good agreement between simulations and ERA5 records in most regions with $r^2 > 0.5$, though
449 degradation was found in a few catchments. Note that, the poor performances were generally found
450 in catchments with limited snow accumulation, e.g., W14-17 in subplots in Figure 4. In other words,
451 the absolute deviation was relatively limited for snow depth simulation.

452

453 **4 Water age simulation and its link to water quality**

454 Like many existing distributed hydrological and water quality models (e.g., SWAT, mHM, Ech₂O-iso,
455 HYPE etc.), EcoTWIN can provide estimation of the main ecohydrological fluxes at high spatial and
456 temporal resolutions, including canopy interception, snow melt-accumulation, infiltration, percolation
457 through soil layers, groundwater recharge, and lateral flux routing at different horizontal phrases.
458 Among these variables, a unique trait of EcoTWIN lies in its capacity to track water fluxes via isotopes,
459 thus being able to provide a consistent estimate of water age and travel times. Therefore, in Figure 5,
460 both variables are shown as the long-term average from 1982 to 2024 for soil profile and stream water.

461 Generally, the magnitudes of water ages follow the geographic and climatic gradients, with younger
462 water found in catchments with higher annual precipitation inputs. Those regions locate in the north-
463 west coast of Europe (Figure 5), particularly for Nordic catchments where lower temperature and net
464 radiation further limit the level of potential evapotranspiration, leading to larger percolation to deeper
465 soil layers and groundwater. Such high turnover rates of water in these catchments (W1, W2, W3, W4,
466 W5, and W8) are also demonstrated as the simulated travel time in soil profile with average values
467 remaining below 500 days.

468 A similar pattern was also found in mountainous regions with higher precipitation and lower potential
469 evapotranspiration compared to lowland areas. Two clear examples are W12 and W17 located in the
470 Alps and the Taurus Mountains where water ages and travel time remained below 500 days (Figure

471 5). In specific wet periods, the water ages and travel time can be reduced to just days, suggesting the
472 rapid response of saturated hydrological systems (e.g., the wet year 1999 in Europe in Figure S5-7). In
473 contrast, the lowlands in central-west Europe showed much slower turnover rates, with the mean
474 water ages reaching almost 10 years in some specific regions. A few examples could be found in the
475 three major representative catchments in Central Europe – Elbe, Rhine, and Danube (W9-11). Such
476 old water ages and long travel time are further exacerbated during dry years (e.g., 2004, a drought
477 year for much of Europe shown in Figure S5-7).

478 Note that though water ages and travel time share similar magnitudes and spatial patterns. It is partly
479 attributed to the fact that the travel time in the conceptualised storages increases exponentially in a
480 sequential order. Taking the Rhine as an example, the average travel time in top soil layer, median soil
481 layer, deep soil layer are 65, 225, 1291 days, respectively. Such a depth-dependence profile makes the
482 overall ages/travel time follow the magnitude of bottom layer and leads to similarity between water
483 ages and travel time. However, large discrepancies are possible between the two indices if a shallow
484 lower boundary is adopted.

485 The estimation of travel time and water ages further provides opportunities to link hydrology and
486 water quality processes in the modelling framework. The simplest and most intuitive way is to
487 compare travel times and simulated biogeochemical process kinetics. Taking denitrification as an
488 example, we applied linear regression and Spearman's correlation test to investigate the potential
489 correlation between travel time of soil water and denitrification rates. The results in Figure 6 showed
490 the strong positive correlations in most agricultural-dominated catchments (W5, W5, W6, W8, W9,
491 W11) yet only weak or no correlation in remaining pristine watersheds. This suggests that travel time
492 might be a key control on soil nitrogen removal in European croplands.

493 More insights can be gained via examination of the Damköhler Number, which quantifies the ratio
494 between timescales of solute transport and biogeochemical transformation. Here in our modelling
495 framework, it can be calculated as the ratio between the travel time of soil water and the time for all
496 soil $\text{NO}_3\text{-N}$ storage to be removed under the simulated denitrification rates. Damköhler numbers <1
497 mean that soil water nitrogen cannot be fully removed during time of residence, indicating the
498 dominance of transport over removal processes and the potential of nitrogen leaching. As shown in
499 Figure 7c, the long-term averages of Damköhler number remain below 1 in most croplands, supporting
500 the conclusion from the linear regression (travel time is a major limiting factor on soil nitrogen
501 removal). Via the spatial- and temporal-explicit estimation of Damköhler number, EcoTWIN provides
502 the opportunity to bridge the catchment hydrological and water quality with travel time.

503

504 **5 Discussion**

505 **5.1 Structural and Functional Merits of EcoTWIN**

506 As a new tracer-aided ecohydrological model, EcoTWIN has novel advantages compared to previous
507 models. In this section, we briefly introduced the merits in model structure, applicability, and insights
508 from tracer-aided simulation.

509 ***5.1.1 Integrated C++ framework***

510 Applications of large-scale modelling have been increasingly popular due to the accelerating
511 development of observation networks and availability of remote sensed data. However, it severely
512 increases the computational burden of ecohydrological modelling. Especially for fully distributed
513 models, increasing size of the model domain can lead to exponential increase in computation demands.
514 In this context, an integrated framework in C++ can significantly accelerate the modelling tasks, as all
515 computation can be conducted within memory thereby avoiding the additional input/output
516 overhead associated with disk-based operations in loosely coupled model chains (e.g., ECH₂O-iso-
517 nitrate; Yang et al., 2024). A standard test was not performed, but based on our modelling experience
518 in the same catchment with different models, the speed of EcoTWIN (~5 seconds for a simulation with
519 285 grid cells and 30 years at daily timestep) is close to the water quality model mHM-Nitrate (~5
520 seconds yet without isotopic simulations; Wu et al., 2022) and easily outperforms ECH₂O-iso-nitrate (7
521 minutes; Wu et al., 2025b).

522 ***5.1.2 Selective disassembly structure***

523 EcoTWIN incorporates a wide range of ecohydrological processes from canopy to groundwater, which
524 not only include natural processes but also anthropogenic activities like irrigation. Land managements
525 can also be represented by dynamic parametrisation, thus enabling EcoTWIN to function as a learning
526 tool to investigate the impacts of changes in anthropogenic management over natural ecosystems;
527 for instance, the land use distribution was updated every 10 years in our test examples to reflect the
528 moderate increases in afforestation in the past 45 years in Europe. More importantly, unlike hard
529 coded process representations/equations in most ecohydrological models, EcoTWIN has a selective
530 disassembly structure, which provides alternative conceptualisations for several important hydrological
531 processes (canopy interception, percolation, groundwater recharge, as well as three pedotransfer
532 functions for initialising soil properties). Modellers can benefit from such flexible model structures by
533 either selecting process representations best suited to field knowledge or data prior to calibration, or
534 integrating module selection into the calibration thus enabling simultaneous optimisation of model
535 structure and corresponding parameters. The latter aspect, i.e., the optimisation of model structure,
536 can be realised together with the recently developed optimisation algorithm DREAM_(LOAX) that aims to
537 identify the deficits in model structure during calibration (Wu et al., 2025a).

538 ***5.1.3 Transferability to contrasting geographic and climatic contexts***

539 To thoroughly test the applicability of EcoTWIN, 17 catchments with different climatic and
540 geographical contexts were selected for calibration and validation, spanning over most biomes in

541 Europe, from snow-dominated watersheds in Nordic or alpine regions, to agricultural-influenced
542 lowlands catchments, and Mediterranean ecosystems (Figure 2 and Table 1). Through multi-criteria
543 calibration against three objectives at multiple sites, the model successfully reproduced the
544 seasonality and peaks of discharge, in-stream isotopes, and $\text{NO}_3\text{-N}$ concentrations in most catchments.
545 Such performance is comparable or better than the previous model benchmarks at similar scales
546 (Bajracharya et al., 2023; Mikayilov et al., 2015; Rakovec et al., 2016, 2019). Note that the
547 concentration of $\text{NO}_3\text{-N}$ was used for calibration, whose accurate simulation is more difficult than $\text{NO}_3\text{-}$
548 N loads given the naturally good performance in discharge. In the other words, hydrological simulation
549 is often the least problematic part in integrated water quality modelling, as it is mostly dominated by
550 natural catchment properties while nitrogen cycling is more interfered by anthropogenic
551 managements (e.g., fertilization and irrigation) (Wu et al., 2025b). Additionally, the simulated internal
552 fluxes were also compared to three reanalysis products in hydrological simulations, corresponding to
553 the key fluxes or storage states in hydrological cycling (snow melt-accumulation, evapotranspiration,
554 and water storage). The results show that constrained by isotopes, EcoTWIN was able to reproduce
555 comparable hydrological modelling results to the remote sensing data without direct calibration
556 regarding magnitudes, spatial patterns, and temporal dynamics. The only degraded performance was
557 found in GRACE surface mass anomaly in coastal regions. There are two potential reasons: (i) the
558 coarse resolution of GRACE might account for mass shifts in both ocean and land, yet EcoTWIN only
559 produces mass anomaly in terrestrial systems; (ii) bidirectional fluxes across the land-ocean interface
560 might drive key changes in coastal systems, which is not considered in current version of EcoTWIN.
561 Nonetheless, given the relatively good agreement with most available data, we conclude that
562 EcoTWIN is applicable across a range of terrestrial ecosystems from boreal to temperate and
563 subtropical climate.

564 ***5.1.4 Bridging hydrology and water quality with water ages***

565 Further to the inference of hydrological and nitrogen processes that is also available in other
566 distributed water quality models (Wellen et al., 2015), a unique trait of EcoTWIN lies in its capacity to
567 track water fluxes and ages with stable water isotopes. As a tracer-aided model, EcoTWIN not only
568 simulates the celerity of catchment response, but tracks the velocity of water via different flow paths.
569 The importance of delineating flow paths within catchments has long been recognized by hydrologists,
570 and has motivated the development of many indices to describe the movement of water molecule at
571 catchment-scale and estimate associated timescales (Sprenger et al., 2019). A few examples are water
572 ages, transient time distribution, and young water fractions (Benettin et al., 2015; Hrachowitz et al.,
573 2013; Jasechko et al., 2016). However, those indices are mainly calculated in a lumped manner where
574 different flow paths in the catchment are characterised as a black box, thus characterising the overall
575 input-output dynamics yet potentially omitting important spatio-temporal variability of hydrological
576 boundary conditions. Instead, EcoTWIN, benefiting from the gridded-based structure, can utilise the
577 increasingly available spatial information (e.g., gridded remote sensing datasets) thus characterising
578 the water ages and travel time in a spatially-explicit manner. Note that simulations of water age/travel
579 time, like other ecohydrological processes, are sensitive to spatial resolution. The coarse resolution

580 used for large catchments (e.g., 5 km in this study) may obscure the sub-grid heterogeneity. For
581 instance, local hydrological hotspots characterized by short travel times and young water ages can be
582 damped or averaged out at coarser resolutions, as reported in modelling studies using ECH2O-iso
583 (Smith et al., 2021; Yang et al., 2023b). However, this limitation can be mitigated by increasing spatial
584 resolution, and it does not undermine the utility of EcoTWIN for water-tracking.

585 Compared to water age which quantifies the age of water within the overall system, travel time,
586 accounting for the water age within a specific storage, is more important in understanding the links
587 between hydrological and nutrient cycles. Such an index, also known as transit time or exposure time,
588 forms one of the fundamental components of water quality modelling. Therefore, the travel time
589 estimated by EcoTWIN has potential to improve the simulation of biogeochemical transformations in
590 water quality models interfaced with simplified hydrological modules (e.g., MONERIS; Bonchkovsky
591 & Osadcha, 2024). Moreover, travel time can be used as a proxy to bridge hydrological processes and
592 biogeochemical transformations. Here we presented a simple framework to calculate the Damköhler
593 Number for denitrification. By using the simulated travel time and reaction timescale (i.e., the time
594 for full removal of nitrogen storage under current denitrification rates), estimation of Damköhler
595 Number was achieved in a spatially- and temporally-explicit manner (Figure 7), which can highlight
596 where and when soil nitrogen removal is constrained by the limited exposure time in the catchment.
597 Such high-resolution information is unique, as the use of this index has been largely restricted to
598 steady-state groundwater systems or riparian/hyporheic zones due to the difficulty in quantifying
599 processing time and residence time at larger scales (Ocampo et al., 2006; Wu et al., 2022).

600

601 **5.2 Limitations and roadmap for future development**

602 Despite these advances, EcoTWIN has limitations. In this section, the uncertainties in model structure
603 and conceptualisation are introduced, as well as the potential roadmaps for future developments.

604 ***5.2.1 Potential towards physics-based conceptualisation of groundwater***

605 Groundwater in EcoTWIN is characterised as two conceptual storages linking with adjacent upstream
606 and downstream storages following the topographic gradients. Such conceptualisation, although has
607 been widely employed in hydrological models (e.g., SWAT, mHM, ECH2O, STARR, etc.), does not align
608 with the physical mechanisms of groundwater routing, as groundwater flow direction follows the
609 hydraulic gradients which may not entirely coincide with topographic gradients (Condon et al., 2021).
610 Such simplified routing has less effect in large catchments with clear topographic gradients (e.g., Rhine
611 starting from Alps to North plain), yet might cause biased estimation in water mass balance for flatter
612 headwater catchments (Yang et al., 2025). Therefore, we plan to further incorporate an additional
613 groundwater module to realise physics-based routing following Darcy's Law in future.

614 ***5.2.2 Revisiting mixing strategies***

615 Mixing strategy is a key component in water quality or tracer models describing the flux-storage
616 behaviours along specific flow paths. There has long been a debate on different mixing assumptions

617 and theories. A typical example is the two-water-world hypothesis, where water storage in the soil
618 profile is differentiated into a tightly-bound pool and a mobile-water pool (McDonnell, 2014). Such
619 conceptualisation is close to the definition of soil matrix flow and preferential flow: the existence of
620 free-flowing preferential flow will bypass the soil matrix vertically and accelerate the lateral drainage
621 via direct connection with channel network (Hrachowitz et al., 2013; Sprenger et al., 2019). However,
622 a complete mixing strategy is often regarded as a reasonable first approximation in many situations
623 and is used in most water quality and tracer models (Jung et al., 2025). This is not only attributed to
624 its computational simplicity, but also the difficulty in conceptualising preferential flow in an
625 evidenced-based manner. In the other words, even with the recognition of preferential flow, its
626 calculation is often hindered by the subsurface heterogeneity in soils and bedrock; a good visualisation
627 is given in Figure 7 in Sprenger et al., (2019). Alternatively, partial mixing has been developed for
628 ecohydrological models (e.g., Hrachowitz et al., 2013), which could be added as a complementary
629 mixing strategy in EcoTWIN. However, as benchmarked in Hrachowitz et al., (2013), the partial mixing
630 brings only moderate improvements in simulations yet can introduce challenges to model spin-up (the
631 increasing instability of storage ages due to the exchange between bypass and storage compartment).
632 Moreover, the realisation of partial mixing, like preferential flow, relies on additional parameters to
633 describe the timing and extent of mixing thus introducing additional parametric uncertainty.
634 Therefore, we recommend a rigorous evaluation of the necessity of partial mixing before any
635 application.

636 ***5.2.3 Complementing the in-stream biogeochemical processes***

637 Transformation is as crucial as transport in inland-water nitrogen cycling (Wang et al., 2024). In the
638 current version of EcoTWIN, denitrification is the only in-stream process of nitrogen loss. However,
639 recent studies have shown that other processes are involved which may be important for aquatic
640 nitrogen cycling. An example originates from Wang et al., (2024), where global inland-water modelling
641 shows that in-stream denitrification only accounts for a minor fraction of $\text{NO}_3\text{-N}$ removal compared to
642 biological uptake. Though their modelling considers lakes and reservoirs where primary production of
643 benthic plants and algae is usually greater than that in rivers, in-stream assimilation might still play a
644 significant role, particularly, in slow-flowing river systems. This is supported by a recent modelling
645 study that estimated nitrogen retention at 15-min interval based on high-frequency $\text{NO}_3\text{-N}$ data (Yang
646 et al., 2023a). Therefore, we plan to further compliment EcoTWIN with in-stream assimilation
647 conceptualisation, as well as other potentially important riverine processes (e.g., nitrogen burial in
648 sediments; Akbarzadeh et al., 2019).

649 ***5.2.4 Integrated calibration framework to embrace equifinality***

650 Strictly speaking, equifinality is not specifically linked to EcoTWIN, but remains a universal problem for
651 calibration or parameter tuning for almost all ecohydrological models. It is reflected in multiple
652 parameters sets yielding similarly good model performance, thus increasing the uncertainty in process
653 inference. The extent of equifinality is primarily controlled by the magnitude of parameters and
654 observation/objectives (Wu et al., 2025c). Unfortunately, conceptualisations across diverse process

655 domains (e.g. for hydrology, isotopes and N-cycling) in EcoTWIN also lead to a relatively large number
656 of parameters. Such risk in equifinality can be potentially constrained via sensitivity analysis, but can
657 still remain an issue given the ubiquitous epistemic uncertainty in data and model structure (Beven,
658 2006, 2015). Alternatively, the recently developed calibration algorithm DREAM_(LoAX) provides an
659 opportunity to embrace equifinality by tuning parameters based on the limits-of-acceptability theory
660 under the equifinality thesis (Wu et al., 2025a). The integrated modelling framework of EcoTWIN and
661 DREAM_(LoAX) can potentially increase the robustness of model calibration and inference.

662

663 **6 Conclusions**

664 Uncertainty is a central concern in ecohydrological modelling, as models are not only used for
665 prediction of specific variables, but also for process inference (backtracking internal processes from
666 available observations) that are inherently embedded within considerable uncertainty. Stable water
667 isotopes can help effectively constrain hydrological fluxes due to their conservative nature, motivating
668 the increased development of tracer-aided models. However, few attempts have been made to
669 incorporate a tracer-aided hydrological framework into water quality models.

670 Therefore, we introduced EcoTWIN, a fully distributed tracer-aided **ecohydrological** model that **tracks**
671 **water**, **isotopic**, and **nutrient** fluxes simultaneously in an integrated C++-based framework. To
672 thoroughly validate the model, 17 large European catchments were selected with a wide range of
673 geographic and climatic gradients (from snow-dominated watersheds in Nordic or alpine regions, to
674 agricultural-influenced lowlands catchments, and Mediterranean ecosystems). The model was
675 calibrated against long-term observations of discharge, in-stream isotopes, and NO₃-N concentrations
676 during 1980-2024 in each of the 17 catchments. Additionally, uncalibrated internal states and fluxes
677 were also compared with three remote sensing products (ERA5 snow depth, MODIS
678 evapotranspiration, and GRACE surface water anomaly) to validate the credibility of process inference.

679 The generally good agreements in both calibrated in-stream components and uncalibrated internal
680 flux-states demonstrated that EcoTWIN is a transferable, flexible prediction and learning tool for
681 process inference across biomes ranging from boreal to subtropical climate. Constrained by tracer
682 simulations, the model not only reproduces the celerity of hydrological systems, but also tracks the
683 velocity. Water ages and travel time are embedded in EcoTWIN to provides spatio-temporal-explicit
684 insights into *when*, *where*, and *how* water moves in the system. Such indices further provide the
685 opportunities to efficiently bridge hydrology and water quality at large catchment-scales. An example
686 was presented using the Damköhler Number to identify regions where denitrification was limited by
687 fast turnover rates of water.

688 Following this “proof of concept” we also see numerous areas where future developments can
689 improve the limitations in the 1.0 version of the model.

690

691 **Code and data availability**

692 The initial version (v1.0) of EcoTWIN is archived in <https://doi.org/10.5281/zenodo.16747633> (Wu et
693 al., 2025d). For further development please refer to GitHub repository: [https://github.com/songjun-](https://github.com/songjunwu/EcoTWIN)
694 [wu/EcoTWIN](https://github.com/songjunwu/EcoTWIN). The geographic data were acquired from Catchment Characterisation and Modelling
695 database (CCM2, version 2.1). The climatic forcing was acquired from E-OBS database
696 (<https://www.ecad.eu/download/ensembles/ensembles.php>). The LAI were acquired from MODIS
697 database (<http://doi.org/10.5067/MODIS/MOD15A2H.006>). Long-term observation of discharge was
698 acquired from GRDC (<https://grdc.bafg.de/>); in-stream isotopic observations were available from
699 Wateriso database (<https://wateriso.utah.edu/waterisotopes/index.html>) and GNIR database
700 (<https://www.iaea.org/services/networks/gnir>); In-stream NO₃-N concentration were acquired from
701 global water quality database, GEMStat (<https://gemstat.org/>).

702

703 **Acknowledgements**

704 Tetzlaff's and Wu's contributions were partly funded through the WETSCAPES2.0 project (DFG
705 TRR410/1 2025) and the Einstein Research Unit "Climate and Water under Change" from the Einstein
706 Foundation Berlin and Berlin University Alliance (grant no. ERU-2020-609). Tetzlaff was also partly
707 funded through Leibniz Excellence project ISOSCALE and received funding from the
708 "Wasserressourcenpreis 2024" of the Rüdiger Kurt Bode-Foundation. This research was also
709 supported by the BMBF (funding code 033W034A), which supported the IGB stable isotope Laboratory.
710 Contributions from Soulsby were supported by Leibnitz Association Germany in the project Wetland
711 Restoration in Peatlands.

712

713 **Author contribution**

714 Conceptualization: SW, DT, YZ, CS

715 Data curation: SW

716 Methodology: SW

717 Software: SW

718 Investigation: SW, DT, YZ, CS

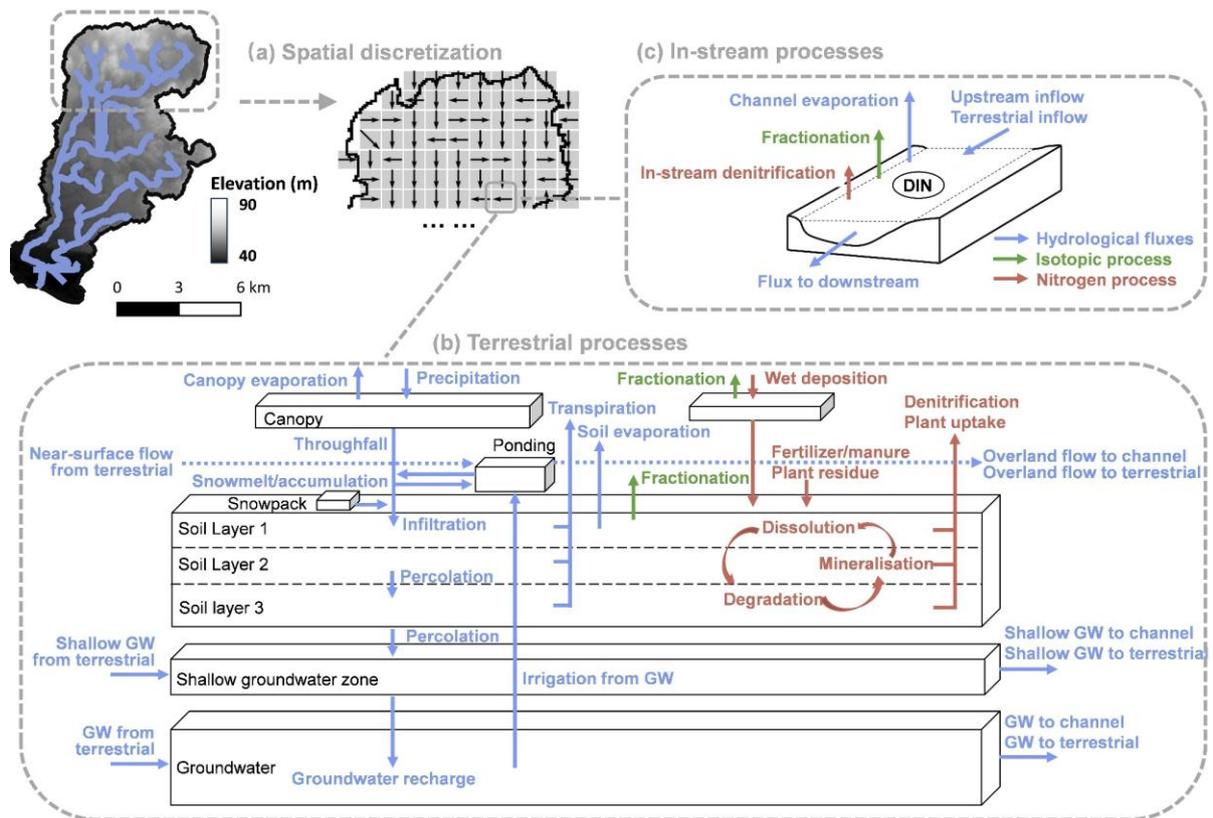
719 Visualization: SW

720 Supervision: DT, CS

721 Writing (original draft preparation): SW

722 Writing (review and editing): SW, DT, YZ, CS

723 **Figures**

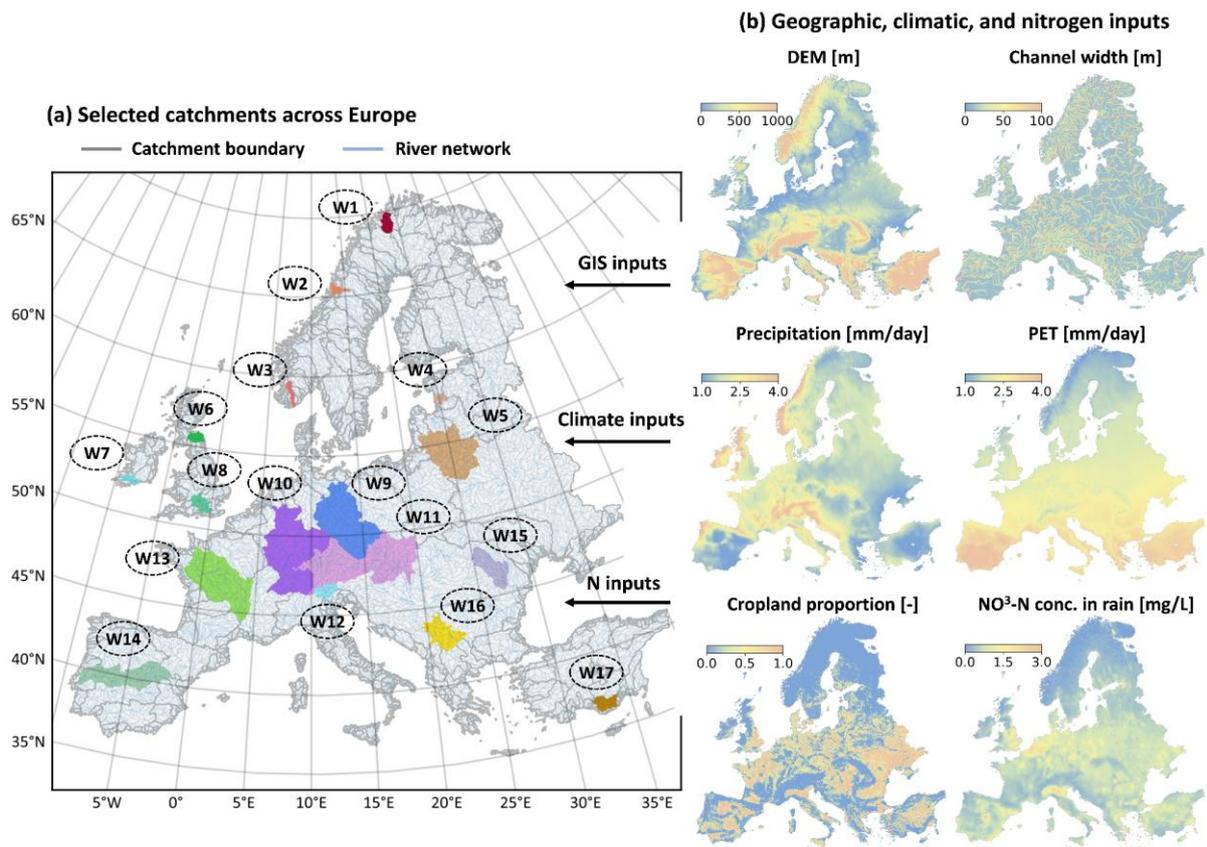


724

725 Figure 1. Model structure of EcoTWIN. As a distributed model, EcoTWIN disentangles the spatial
 726 domain into grid cells (Panel a). In each grid cell, hydrological, isotopic, and nitrogen processes were
 727 simulated in canopy, snow, soils, shallow groundwater, and groundwater (panel b) and river channel
 728 if channels are present (Panel c).

729

730

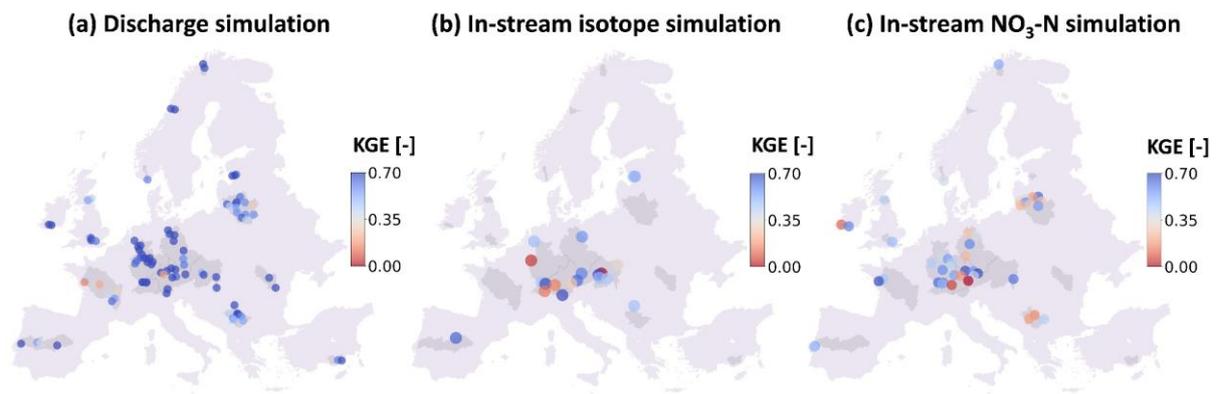


731

732 Figure 2. The selected catchments for model validation (Panel a) and an overview of key geographic,
733 climatic, and nitrogen inputs (Panel b).

734

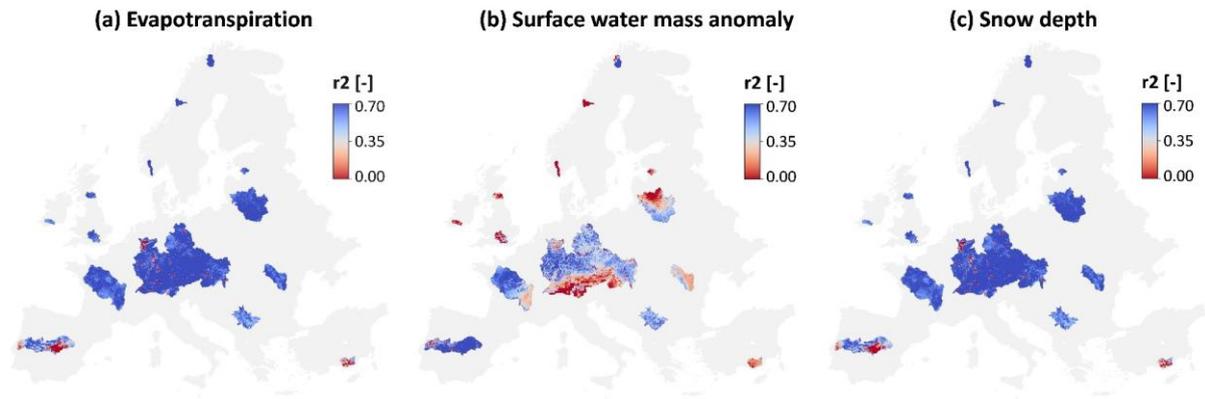
735



736

737 Figure 3. The simulation performance of discharge, in-stream isotope, and in-stream NO₃-N. The
738 simulated and observed time series are shown in Figure S1.

739

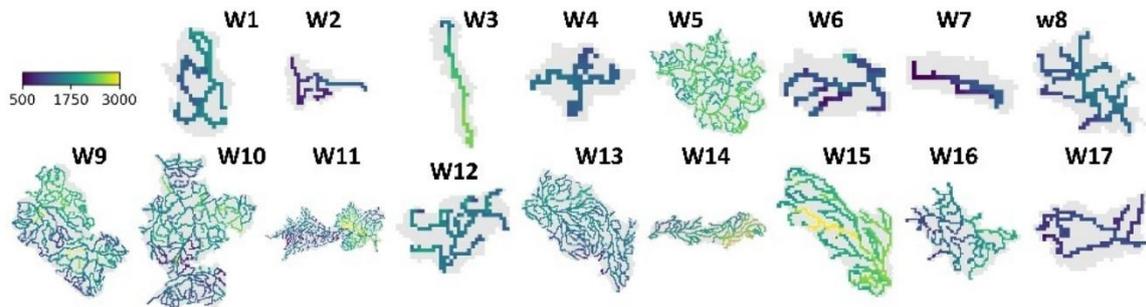


740

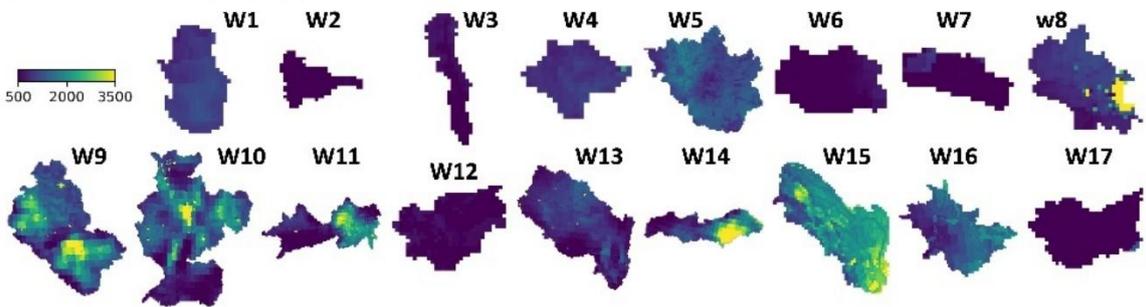
741 Figure 4. The grid-to-grid comparison between time series of simulated internal states/fluxes and the
 742 ones extracted from remote sensing/reanalysis products, including evapotranspiration from MODIS
 743 (Panel a), surface water mass anomaly from GRACE (Panel b), and snow depth from ERA5 (Panel c).
 744 The time series of simulation and remote sensing/reanalysis products in each catchment are shown
 745 Figure S2-4.

746

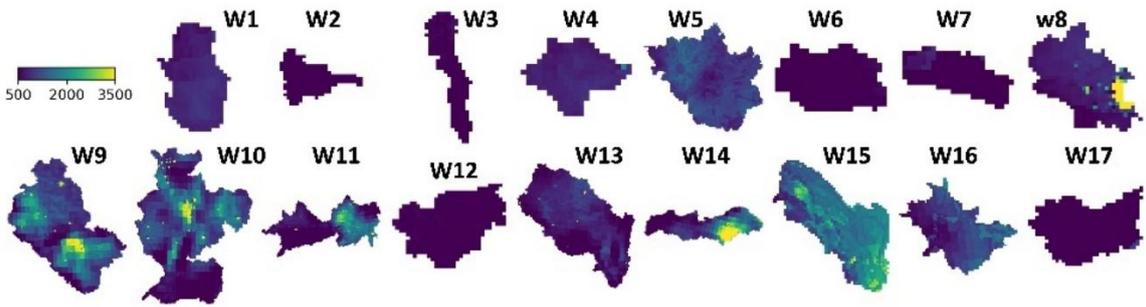
(a) Stream water age [days]



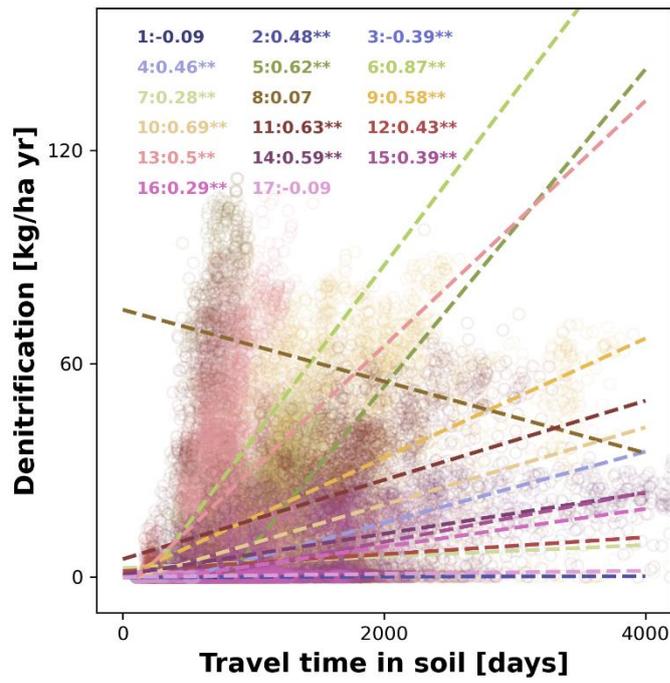
(b) Soil water ages [days]



(c) Travel time in soils [days]



749 Figure 5. The simulated long-term average (1982-2024) of water age and travel time in channel and
750 soil profile. Water ages represent the time since water enters the catchments as precipitation, while
751 travel times depict the residence time of water within the specific storage.

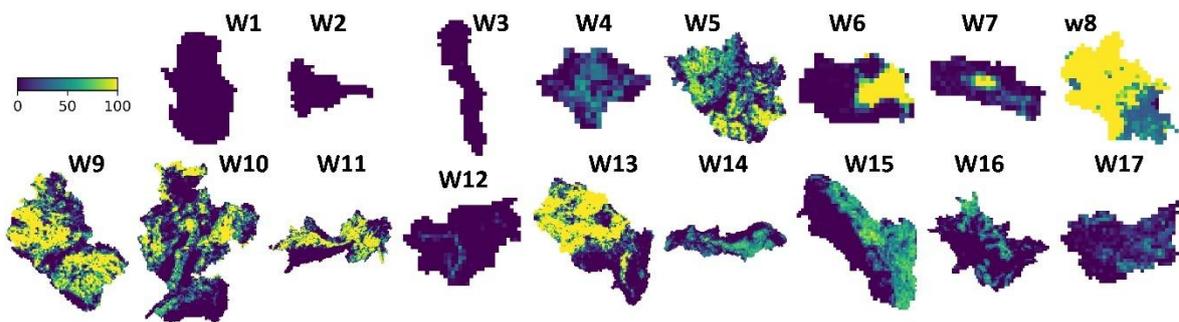


753

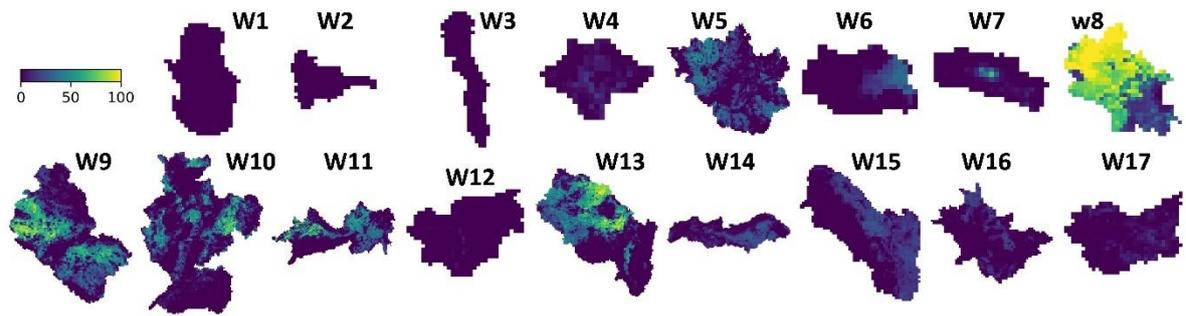
754 Figure 6. The correlations between travel time and annual denitrification. The text depicts the
 755 spearman correlation coefficients and p values (* = less than 0.05, ** = less than 0.01) in each
 756 catchment.

757

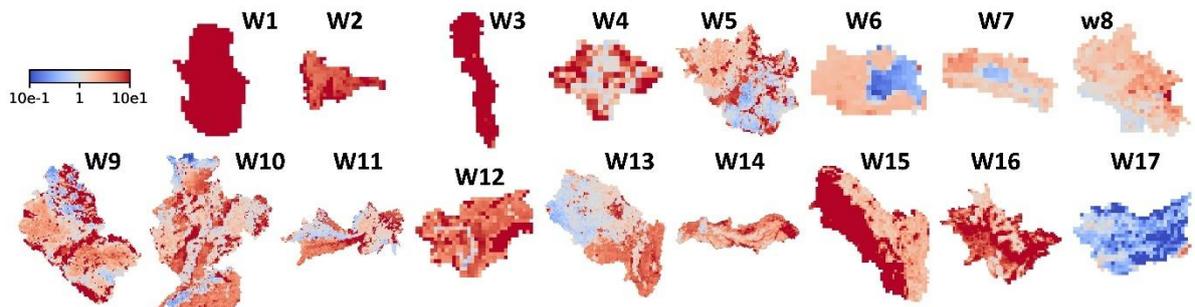
(a) Nitrogen inputs [kg/ha yr]



(b) Soil denitrification [kg/ha yr]



(c) Damköhler number [-]



758

759 Figure 7. The simulated long-term average (1982-2024) of nitrogen inputs, soil denitrification, and
760 Damköhler number.

761

762

763 **Tables**

764 Table 1. Characteristics of the selected catchments. **Lat** depicts the latitude of upper left corner of the
 765 catchment. **DEM** and **Area** are the mean elevation in m.a.s.l. and catchment size in km². **Precip**, **Temp**,
 766 and **PET** are the annual averages of precipitation, air temperature, and potential evapotranspiration
 767 in mm/yr. f_{crop} , f_{forest} , and f_{urban} are the fractions of cropland, forest, and urbanized areas in 2019
 768 in %. Null means no name is assigned for the catchment in the Catchment Characterisation and
 769 Modelling (CCM) database.

ID	Name	Lat	Area	DEM	Precip	Temp	PET	f_{crop}	f_{forest}	f_{urban}
1	Null	70.0	8725	468.5	448.5	-1.8	442.8	<1	1.9	<1
2	Vefsna	65.9	5475	636.5	1260.8	0.7	433.6	<1	24.6	<1
3	Null	59.8	5225	742.3	1400.9	3.0	545.7	<1	39.8	<1
4	Null	58.3	4350	67.4	654.5	6.4	667.9	17.4	60.5	1.4
5	Nemunas	56.6	97550	147.9	599.3	7.1	730.6	33.0	39.1	4.5
6	Tweed	55.9	6250	264.3	1023.4	7.9	600.9	21.8	18.9	1.4
7	Null	52.3	4300	175.4	1218.1	10.1	645.2	12.1	17.4	1.9
8	Thames	52.2	11900	112.0	700.7	10.4	782.9	44.9	14.1	22.0
9	Elbe	53.5	130225	318.3	626.9	8.8	836.3	41.2	34.6	10.8
10	Rhine	52.0	170175	508.3	943.3	8.9	821.0	21.5	41.2	17.1
11	Danube ^(a)	50.5	197600	618.0	843.6	8.3	857.4	28.5	37.1	11.5
12	Adige	47.2	11600	1771.9	1002.3	4.5	809.7	<1	48.8	3.2
13	Loire	48.7	122125	298.9	778.7	11.0	887.4	37.6	25.7	6.5
14	Tajo	40.4	75575	686.2	549.5	14.3	1359.6	26.7	34.5	3.3
15	Danube ^(b)	48.4	37975	533.3	534.7	8.1	869.5	32.0	41.3	6.4
16	Danube ^(c)	44.8	37725	653.3	684.7	9.7	994.2	12.8	44.3	5.8
17	Null	37.6	12650	1384.5	454.1	12.2	1256.9	5.9	4.3	<1

770

771

772 Table 2. The calibration performance of discharge (Q), in-stream isotopes (¹⁸O, Iso), and nitrate (NO³-
 773 N). Evaluation metrics include Kling-Gupta efficiency (-), Root Mean Square Error (m³/s, ‰, and mg/L
 774 for discharge, isotopes, and nitrate, respectively), Pearson Correlation Coefficient (-), and Percent bias
 775 (%).

Metric	Unit	Min	Max	Mean	Median
Kling-Gupta efficiency (Q)	-	0.14	0.89	0.65	0.69
Kling-Gupta efficiency (Iso)	-	-0.03	0.86	0.45	0.48
Kling-Gupta efficiency (NO ³ -N)	-	-0.36	0.72	0.42	0.44
Pearson correlation coefficient (Q)	-	0.49	0.92	0.79	0.81
Pearson correlation coefficient (Iso)	-	0.14	0.87	0.51	0.54
Pearson correlation coefficient (NO ³ -N)	-	-0.26	0.86	0.55	0.6
Root Mean Square Error (Q)	m ³ /s	3.99	677.08	123.02	68.51
Root Mean Square Error (Iso)	‰	0.31	1.51	0.72	0.73
Root Mean Square Error (NO ³ -N)	mg/L	0.02	2.82	0.83	0.57
Percent bias (Q)	%	0.52	79.88	17.44	9.53
Percent bias (Iso)	%	-11.28	-0.07	-4.3	-4.42
Percent bias (NO ³ -N)	%	0.18	49.25	15.52	10.89

776

777 **Reference**

- 778 Akbarzadeh, Z., Maavara, T., Slowinski, S., & Van Cappellen, P. (2019). Effects of Damming on River
779 Nitrogen Fluxes: A Global Analysis. *Global Biogeochemical Cycles*, 33.
780 <https://doi.org/10.1029/2019GB006222>
- 781 Ala-aho, P., Tetzlaff, D., Mcnamara, J., Laudon, H., & Kormos, P. (2017). Modeling the isotopic
782 evolution of snowpack and snowmelt: Testing a spatially distributed parsimonious approach.
783 *Water Resources Research*, 53. <https://doi.org/10.1002/2017WR020650>
- 784 Amato, M. T., & Giménez, D. (2024). Predicting monthly near-surface soil temperature from air
785 temperature and the leaf area index. *Agricultural and Forest Meteorology*, 345, 109838.
786 <https://doi.org/10.1016/j.agrformet.2023.109838>
- 787 Arnold, J., Moriasi, D., Gassman, P., Abbaspour, K., White, M., Srinivasan, R., et al. (2012). SWAT:
788 Model use, calibration, and validation. *Transactions of the ASABE (American Society of*
789 *Agricultural and Biological Engineers)*, 55. <https://doi.org/10.13031/2013.42256>
- 790 Bajracharya, A., Moghairib, M., Stadyk, T., & Asadzadeh, M. (2023). Process based calibration of a
791 continental-scale hydrological model using soil moisture and streamflow data. *Journal of*
792 *Hydrology: Regional Studies*, 47, 101391. <https://doi.org/10.1016/j.ejrh.2023.101391>
- 793 Benettin, P., Bailey, S., Campbell, J., Green, M., Rinaldo, A., Likens, G., et al. (2015). Linking water age
794 and solute dynamics in streamflow at the Hubbard Brook Experimental Forest, NH, USA.
795 *Water Resources Research*, 51, 9256–9272. <https://doi.org/10.1002/2015WR017552>
- 796 Beven, K. (2006). A manifesto for the equifinality thesis. *Journal of Hydrology*, 320(1–2), 18–36.
797 <https://doi.org/10.1016/J.JHYDROL.2005.07.007>
- 798 Beven, K. (2015). Facets of uncertainty: Epistemic uncertainty, non-stationarity, likelihood, hypothesis
799 testing, and communication. *Hydrological Sciences Journal*, 61, 150407155940006.
800 <https://doi.org/10.1080/02626667.2015.1031761>
- 801 Birkel, C., & Soulsby, C. (2015). Advancing tracer-aided rainfall-runoff modelling: A review of progress,
802 problems and unrealised potential. *Hydrological Processes*, 29(25), 5227–5240.
803 <https://doi.org/10.1002/HYP.10594>
- 804 Birkel, C., Tetzlaff, D., & Dunn, S. (2011). Using time domain and geographic source tracers to
805 conceptualize streamflow generation processes in lumped rainfall-runoff models. *Water*
806 *Resources Research - WATER RESOUR RES*, 47. <https://doi.org/10.1029/2010WR009547>
- 807 Bonchkovskiy, A., & Osadcha, N. (2024). Modelling of the nutrient load in the Sula River basin using
808 the MONERIS. *Physical Geography and Geomorphology*, 47, 7–20.
809 <https://doi.org/10.17721/phgg.2024.3-4.01>
- 810 Condon, L., Kollet, S., Bierkens, M., Maxwell, R., Hill, M., Franssen, H.-J., et al. (2021). Global
811 Groundwater Modeling and Monitoring: Opportunities and Challenges. *Water Resources*
812 *Research*, 57. <https://doi.org/10.1029/2020WR029500>
- 813 Craig, H., Gordon, L. I., & Horibe, Y. (1964). Isotopic exchange effects in the evaporation of water: 1.
814 Low-temperature experimental results. *Journal of Geophysical Research*, 68(17), 5079–5087.
815 <https://doi.org/10.1029/JZ068I017P05079>
- 816 Dawson, T., & Ehleringer, J. (1991). Streamside trees that do not use stream water. *Nature*, 350, 335–
817 337. <https://doi.org/10.1038/350335a0>
- 818 Dick, J. J., Tetzlaff, D., Birkel, C., & Soulsby, C. (2015). Modelling landscape controls on dissolved
819 organic carbon sources and fluxes to streams. *Biogeochemistry*, 122(2–3), 361–374.
820 <https://doi.org/10.1007/S10533-014-0046-3>
- 821 Donnelly, C., Andersson, J. C. M., & Arheimer, B. (2016). Using flow signatures and catchment
822 similarities to evaluate the E-HYPE multi-basin model across Europe. *Hydrological Sciences*
823 *Journal*, 61(2), 255–273. <https://doi.org/10.1080/02626667.2015.1027710>

824 Eckersten, H., Jansson, P. E., & Johnsson, H. (1994). SOILN model – user’s manual 2nd edition, Division
825 of Agricultural Hydrotechnics Communications, 94:4. Department of Soil Sciences, Swedish
826 University of Agricultural Sciences, 58pp, Uppsala.

827 Efstratiadis, A., & Koutsoyiannis, D. (2010). One decade of multi-objective calibration approaches in
828 hydrological modelling: A review. *Hydrological Sciences Journal*, 55(1), 58–78.
829 <https://doi.org/10.1080/02626660903526292>

830 Godsey, S. E., Aas, W., Clair, T. A., de Wit, H. A., Fernandez, I. J., Kahl, J. S., et al. (2010). Generality of
831 fractal 1/f scaling in catchment tracer time series, and its implications for catchment travel
832 time distributions. *Hydrological Processes*, 24(12), 1660–1671.
833 <https://doi.org/10.1002/hyp.7677>

834 Good, S. P., Soderberg, K., Guan, K., King, E. G., Scanlon, T. M., & Caylor, K. K. (2014). $\delta^2\text{H}$ isotopic flux
835 partitioning of evapotranspiration over a grass field following a water pulse and subsequent
836 dry down. *Water Resources Research*, 50(2), 1410–1432.
837 <https://doi.org/10.1002/2013WR014333>

838 Grizzetti, B., Vigiak, O., Udias, A., Aloe, A., Zanni, M., Bouraoui, F., et al. (2021). How EU policies could
839 reduce nutrient pollution in European inland and coastal waters. *Global Environmental*
840 *Change*, 69, 102281. <https://doi.org/10.1016/j.gloenvcha.2021.102281>

841 Hersbach, H., Bell, B., Berrisford, P., Hirahara, S., Horányi, A., Muñoz Sabater, J., et al. (2020). The ERA5
842 global reanalysis. *Quarterly Journal of the Royal Meteorological Society*, 146.
843 <https://doi.org/10.1002/qj.3803>

844 Hooper, R., Stone, A., Christophersen, N., de Melo, E., & Seip, H. (1988). Assessing the Birkenes Model
845 of Stream Acidification using a multi-signal calibration methodology. *Water Resources*
846 *Research - WATER RESOUR RES*, 24, 1308–1316. <https://doi.org/10.1029/WR024i008p01308>

847 Horita, J., Rozanski, K., & Cohen, S. (2008). Isotope effects in the evaporation of water: A status report
848 of the Craig-Gordon model. *Isotopes in Environmental and Health Studies*, 44, 23–49.
849 <https://doi.org/10.1080/10256010801887174>

850 Hrachowitz, M., Savenije, H., Bogaard, T. A., Tetzlaff, D., & Soulsby, C. (2013). What can flux tracking
851 teach us about water age distribution patterns and their temporal dynamics? *Hydrology and*
852 *Earth System Sciences*, 117(2), 533–564. <https://doi.org/10.5194/HESS-17-533-2013>

853 Huijgevoort, M., Tetzlaff, D., & Sutanudjaja, E. (2016). Using high resolution tracer data to constrain
854 water storage, flux and age estimates in a spatially distributed rainfall-runoff model.
855 *Hydrological Processes*, 30. <https://doi.org/10.1002/hyp.10902>

856 Jasechko, S., Kirchner, J., Welker, J., & McDonnell, J. (2016). Substantial proportion of global
857 streamflow less than three months old. *Nature Geoscience*, 9.
858 <https://doi.org/10.1038/NGEO2636>

859 Jones, E. R., Bierkens, M. F. P., Wanders, N., Sutanudjaja, E. H., van Beek, L. P. H., & van Vliet, M. T. H.
860 (2023). DynQual v1.0: a high-resolution global surface water quality model. *Geoscientific*
861 *Model Development*, 16(15), 4481–4500. <https://doi.org/10.5194/gmd-16-4481-2023>

862 Jung, H., Tetzlaff, D., Birkel, C., & Soulsby, C. (2025). Recent Developments and Emerging Challenges
863 in Tracer-Aided Modeling. *Wiley Interdisciplinary Reviews Water*, 12.
864 <https://doi.org/10.1002/wat2.70015>

865 Kale, R., & Sahoo, B. (2011). Green-Ampt Infiltration Models for Varied Field Conditions: A Revisit.
866 *Water Resources Management*, 25, 3505–3536. <https://doi.org/10.1007/s11269-011-9868-0>

867 Kumar, R., Samaniego, L., & Attinger, S. (2013). Implications of distributed hydrologic model
868 parameterization on water fluxes at multiple scales and locations. *Water Resources Research*,
869 49(1), 360–379. <https://doi.org/10.1029/2012WR012195>

- 870 Kuppel, S., Tetzlaff, D., Maneta, M., & Soulsby, C. (2018). EcH2O-iso 1.0: Water isotopes and age
871 tracking in a process-based, distributed ecohydrological model. *Geoscientific Model*
872 *Development*, 11, 3045–3069. <https://doi.org/10.5194/gmd-11-3045-2018>
- 873 Landgraf, J., Tetzlaff, D., Birkel, C., Stevenson, J., & Soulsby, C. (2023). Assessing land use effects on
874 ecohydrological partitioning in the critical zone through isotope-aided modelling. *Earth*
875 *Surface Processes and Landforms*, 48. <https://doi.org/10.1002/esp.5691>
- 876 Lindström, G., Pers, C., Rosberg, J., Strömqvist, J., & Arheimer, B. (2010). Development and test of the
877 HYPE (Hydrological Predictions for the Environment) model – A water quality model for
878 different spatial scales. *Hydrology Research*, 41. <https://doi.org/10.2166/nh.2010.007>
- 879 Maneta, M. P., & Silverman, N. L. (2013). A spatially distributed model to simulate water, energy, and
880 vegetation dynamics using information from regional climate models. *Earth Interactions*,
881 17(11), 1–44. <https://doi.org/10.1175/2012EI000472.1>
- 882 McDonnell, J. (2014). The two water worlds hypothesis: Ecohydrological separation of water between
883 streams and trees? *Wiley Interdisciplinary Reviews: Water*, 1.
884 <https://doi.org/10.1002/wat2.1027>
- 885 McDonnell, J., & Beven, K. (2014). Debates on Water Resources: The future of hydrological sciences:
886 A (common) path forward? A call to action aimed at understanding velocities, celerities and
887 residence time distributions of the headwater hydrograph. *Water Resources Research*, 50.
888 <https://doi.org/10.1002/2013WR015141>
- 889 Mcmillan, H., Araki, R., Bolotin, L., Kim, D.-H., Coxon, G., Clark, M., & Seibert, J. (2025). Global patterns
890 in observed hydrologic processes. *Nature Water*, 3. <https://doi.org/10.1038/s44221-025-00407-w>
- 892 Mikayilov, F., Rouholahnejad Freund, E., Ashraf Vaghefi, S., Srinivasan, R., Yang, H., & Klöve, B. (2015).
893 A continental-scale hydrology and water quality model for Europe: Calibration and uncertainty
894 of a high-resolution large-scale SWAT model. *Journal of Hydrology*, 524.
895 <https://doi.org/10.1016/j.jhydrol.2015.03.027>
- 896 Mu, Q., Zhao, M., & Running, S. (2011). Improvements to a MODIS Global Terrestrial
897 Evapotranspiration Algorithm. *Remote Sensing of Environment*, 115, 1781–1800.
898 <https://doi.org/10.1016/j.rse.2011.02.019>
- 899 Nan, Y., He, Z., Tian, F., Wei, Z., & Tian, L. (2021). Can we use precipitation isotope outputs of isotopic
900 general circulation models to improve hydrological modeling in large mountainous
901 catchments on the Tibetan Plateau? *Hydrology and Earth System Sciences*, 25(12), 6151–6172.
902 <https://doi.org/10.5194/hess-25-6151-2021>
- 903 Naz, B. S., Sharples, W., Ma, Y., Goergen, K., & Kollet, S. (2023). Continental-scale evaluation of a fully
904 distributed coupled land surface and groundwater model, ParFlow-CLM (v3.6.0), over Europe.
905 *Geoscientific Model Development*, 16(6), 1617–1639. <https://doi.org/10.5194/gmd-16-1617-2023>
- 907 Neal, C., Christophersen, N., Neale, R., Smith, C., & Reynolds, B. (1988). Chloride in precipitation and
908 streamwater for the upland catchment of River Severn, mid- Wales; some consequences for
909 hydrochemical models. *Hydrological Processes*, 2, 155–165.
910 <https://doi.org/10.1002/hyp.3360020206>
- 911 Nearing, G., Ruddell, B., Bennett, A., Prieto, C., & Gupta, H. (2020). Does Information Theory Provide
912 a New Paradigm for Earth Science? Hypothesis Testing. *Water Resources Research*, 56.
913 <https://doi.org/10.1029/2019WR024918>
- 914 Ocampo, C., Sivapalan, M., & Oldham, C. (2006). Hydrological connectivity of upland-riparian zones in
915 agricultural catchments: Implications for runoff generation and nitrate transport. *Journal of*
916 *Hydrology*, 331, 643–658. <https://doi.org/10.1016/j.jhydrol.2006.06.010>

917 Pechlivanidis, I., Jackson, B., Mcintyre, N., & Wheeler, H. (2011). Catchment scale hydrological
918 modelling: A review of model types, calibration approaches and uncertainty analysis methods
919 in the context of recent developments in technology and applications. *GlobalNEST*
920 *International Journal*, 13, 193–214.

921 Pesántez, J., Birkel, C., Gaona, G., Arciniega-Esparza, S., Murray, D., Mosquera, G., et al. (2023).
922 Spatially distributed tracer-aided modelling to explore DOC dynamics, hot spots and hot
923 moments in a tropical mountain catchment. *Hydrological Processes*, 37, 1–15.
924 <https://doi.org/10.1002/hyp.15020>

925 Rakovec, O., Kumar, R., Mai, J., Cuntz, M., Thober, S., Zink, M., et al. (2016). Multiscale and Multivariate
926 Evaluation of Water Fluxes and States over European River Basins. *Journal of*
927 *Hydrometeorology*, 17, 287–307. <https://doi.org/10.1175/JHM-D-15-0054.1>

928 Rakovec, O., Mizukami, N., Kumar, R., Newman, A., Thober, S., Wood, A., et al. (2019). Diagnostic
929 Evaluation of Large-Domain Hydrologic Models Calibrated Across the Contiguous United
930 States. *Journal of Geophysical Research: Atmospheres*, 124.
931 <https://doi.org/10.1029/2019JD030767>

932 Remondi, F., Kirchner, J., Burlando, P., & Fatichi, S. (2018). Water Flux Tracking with A Distributed
933 Hydrological Model to Quantify Controls on the Spatio-Temporal Variability of Transit Time
934 Distributions. *Water Resources Research*, 54. <https://doi.org/10.1002/2017WR021689>

935 Samaniego, L., Kumar, R., & Attinger, S. (2010). Multiscale parameter regionalization of a grid-based
936 hydrologic model at the mesoscale. *Water Resour. Res.*, 46.
937 <https://doi.org/10.1029/2008WR007327>

938 Seybold, E., Dwivedi, R., Musselman, K., Kincaid, D., Schroth, A., Classen, A., et al. (2022). Winter runoff
939 events pose an unquantified continental-scale risk of high wintertime nutrient export.
940 *Environmental Research Letters*, 17, 104044. <https://doi.org/10.1088/1748-9326/ac8be5>

941 Šimůnek, J., Šejna, M., Saito, H., & Van Genuchten, M. T. (2013). *The HYDRUS-1D software package*
942 *for simulating the one-dimensional movement of water, heat, and multiple solutes in variably-*
943 *saturated media (Version 4.17)*. Department of Environmental Sciences University of Riverside
944 California, California.

945 Skrzypek, G., Mydłowski, A., Dogramaci, S., Hedley, P., Gibson, J. J., & Grierson, P. F. (2015). Estimation
946 of evaporative loss based on the stable isotope composition of water using Hydrocalculator.
947 *Journal of Hydrology*, 523, 781–789. <https://doi.org/10.1016/J.JHYDROL.2015.02.010>

948 Smith, A., Tetzlaff, D., Kleine, L., Maneta, M., & Soulsby, C. (2021). Quantifying the effects of land use
949 and model scale on water partitioning and water ages using tracer-aided ecohydrological
950 models. *Hydrology and Earth System Sciences*, 25, 2239–2259. [https://doi.org/10.5194/hess-](https://doi.org/10.5194/hess-25-2239-2021)
951 [25-2239-2021](https://doi.org/10.5194/hess-25-2239-2021)

952 Soulsby, C., Birkel, C., Geris, J., Dick, J., Tunaley, C., & Tetzlaff, D. (2015). Stream water age distributions
953 controlled by storage dynamics and nonlinear hydrologic connectivity: Modeling with high-
954 resolution isotope data. *Water Resources Research*, 51(9), 7759–7776.
955 <https://doi.org/10.1002/2015WR017888>

956 Sprenger, M., Stumpp, C., Weiler, M., Aeschbach, W., Allen, S., Benettin, P., et al. (2019). The
957 Demographics of Water: A Review of Water Ages in the Critical Zone. *Reviews of Geophysics*,
958 57. <https://doi.org/10.1029/2018RG000633>

959 Tapley, B., Bettadpur, S., Ries, J. C., Thompson, P., & Watkins, M. (2004). GRACE Measurements of
960 Mass Variability in the Earth System. *Science (New York, N.Y.)*, 305, 503–5.
961 <https://doi.org/10.1126/science.1099192>

962 Tetzlaff, D., Buttle, J., Carey, S. K., Mcguire, K., Laudon, H., & Soulsby, C. (2015). Tracer-based
963 assessment of flow paths, storage and runoff generation in northern catchments: A review.
964 *Hydrological Processes*, 29(16), 3475–3490. <https://doi.org/10.1002/HYP.10412>

- 965 Wang, J., Bouwman, A., Vilmin, L., Beusen, A., Hoek, W., Liu, X., & Middelburg, J. (2024). Global inland-
 966 water nitrogen cycling has accelerated in the Anthropocene. *Nature Water*, 2.
 967 <https://doi.org/10.1038/s44221-024-00282-x>
- 968 Wellen, C., Kamran-Disfani, A.-R., & Arhonditsis, G. (2015). Evaluation of the Current State of
 969 Distributed Watershed Nutrient Water Quality Modeling. *Environmental Science &*
 970 *Technology*, 49. <https://doi.org/10.1021/es5049557>
- 971 Wen, Y., Lin, J.-S., Plaza, F., & Liang, X. (2024). Roles of Hydrology and Transport Processes in
 972 Denitrification at Watershed Scale. *Water Resources Research*, 60, 1–24.
 973 <https://doi.org/10.1029/2023WR034971>
- 974 Winkler, K., Fuchs, R., Rounsevell, M., & Herold, M. (2021). Global land use changes are four times
 975 greater than previously estimated. *Nature Communications*, 12(1), 2501.
 976 <https://doi.org/10.1038/s41467-021-22702-2>
- 977 Wu, S., Tetzlaff, D., Yang, X., & Soulsby, C. (2022). Disentangling the influence of landscape
 978 characteristics, hydroclimatic variability and land management on surface water NO₃-N
 979 dynamics: spatially distributed modelling over 30 years in a lowland mixed land use catchment.
 980 *Water Resources Research*, e2021WR030566. <https://doi.org/10.1029/2021WR030566>
- 981 Wu, S., Tetzlaff, D., Beven, K., & Soulsby, C. (2025a). DREAM(LoAX): Simultaneous Calibration and
 982 Diagnosis for Tracer-Aided Ecohydrological Models Under the Equifinality Thesis. *Water*
 983 *Resources Research*, 61, e2024WR038779. <https://doi.org/10.1029/2024WR038779>
- 984 Wu, S., Tetzlaff, D., Yang, X., Sauter, T., & Soulsby, C. (2025b). Hydrological Connectivity Dominates
 985 NO₃-N Cycling in Complex Landscapes – Insights From Integration of Isotopes and Water
 986 Quality Modeling. *Water Resources Research*, 61, e2025WR040525.
 987 <https://doi.org/10.1029/2025WR040525>
- 988 Wu, S., Tetzlaff, D., & Soulsby, C. (2025c). Revising Common Approaches for Calibration: Insights From
 989 a 1-D Tracer-Aided Hydrological Model With High-Dimensional Parameters and Objectives.
 990 *Water Resources Research*, 61, e2024WR037656. <https://doi.org/10.1029/2024WR037656>
- 991 Wu, S., Tetzlaff, D., Zheng, Y., & Soulsby, C. (2025d). EcoTWIN v1.0 release.
 992 <https://doi.org/10.5281/zenodo.16747633>
- 993 Yang, C., Condon, L., & Maxwell, R. (2025). Unravelling groundwater–stream connections over the
 994 continental United States. *Nature Water*, 3. <https://doi.org/10.1038/s44221-024-00366-8>
- 995 Yang, X., Jomaa, S., Zink, M., Fleckenstein, J., Borchardt, D., & Rode, M. (2018). A New Fully Distributed
 996 Model of Nitrate Transport and Removal at Catchment Scale. *Water Resources Research*, 54.
 997 <https://doi.org/10.1029/2017WR022380>
- 998 Yang, X., Zhang, X., Graeber, D., Hensley, R., Jarvie, H., Lorke, A., et al. (2023a). Large-stream nitrate
 999 retention patterns shift during droughts: Seasonal to sub-daily insights from high-frequency
 1000 data-model fusion. *Water Research*, 243, 120347.
 1001 <https://doi.org/10.1016/j.watres.2023.120347>
- 1002 Yang, X., Tetzlaff, D., Müller, C., Knöller, K., Borchardt, D., & Soulsby, C. (2023b). Upscaling Tracer-
 1003 Aided Ecohydrological Modeling to Larger Catchments: Implications for Process
 1004 Representation and Heterogeneity in Landscape Organization. *Water Resources Research*, 59,
 1005 e2022WR033033. <https://doi.org/10.1029/2022WR033033>
- 1006 Yang, X., Tetzlaff, D., Jin, J., Li, Q., Borchardt, D., & Soulsby, C. (2024). Linking terrestrial biogeochemical
 1007 processes and water ages to catchment water quality: A new Damköhler analysis based on
 1008 coupled modeling of isotope tracers and nitrate dynamics. *Water Research*, 262, 122118.
 1009 <https://doi.org/10.1016/j.watres.2024.122118>
- 1010 Zhang, Z., Chen, X., Cheng, Q.-B., Li, S.-L., Yue, F.-J., Peng, T., et al. (2020). Coupled hydrological and
 1011 biogeochemical modelling of nitrogen transport in the karst critical zone. *Science of The Total*
 1012 *Environment*, 732, 138902. <https://doi.org/10.1016/j.scitotenv.2020.138902>

1013 Zhu, J., Jia, Y., Yu, G., Wang, Q., He, N., Chen, Z., et al. (2025). Changing patterns of global nitrogen
1014 deposition driven by socio-economic development. *Nature Communications*, 16(1), 46.
1015 <https://doi.org/10.1038/s41467-024-55606-y>
1016