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Evaluation of Agricultural Measures to Safeguard the Vulnerable Karst Groundwater Habitat of the Black Olm (*Proteus anguinus parkelj*) from Nitrate Pollution

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Abstract: The black olm (*Proteus anguinus parkelj* Sket & Arntzen) is an endemic species found exclusively in the Dobličica River subterranean water systems of the Dinaric karst in southern Slovenia. These unique habitats are vulnerable to contamination due to rapid water flow, primarily from nitrates from agricultural fertilisers and untreated urban wastewater. The safe limit of nitrate concentration for olms is 9.2 mg NO_3^-/L, yet measurements in karst springs have shown levels ranging from 3 mg to over 20 mg NO_3^-/L. The SWAT modelling tool assessed agri-environmental and land use scenarios for their impact on nitrate leaching. Using the model, we identified hotspots with high nitrogen leaching potential that require immediate attention and implementation of better agricultural practices for fertiliser use. For these hotspots, the most effective approach combines scenarios of cover crops (R2), reduced fertilisation (R3), crop rotation (R4), and conversion of cropland to grassland (E2, E4, E5), potentially decreasing nitrate leaching by up to 60%. Implementing the best scenarios is expected to reduce nitrogen levels below the limit value of 9.2 mg NO_3^-/L, essential for maintaining the black olm habitat.

Keywords: black olm; karst; groundwater; habitat; agriculture; measures; nitrate; SWAT model

Citation: Glavan, M.; Cvejić, R. Evaluation of Agricultural Measures to Safeguard the Vulnerable Karst Groundwater Habitat of the Black Olm (*Proteus anguinus parkelj*) from Nitrate Pollution. *Sustainability* **2024**, *16*, 11309. https://doi.org/10.3390/su162411309

Academic Editors: Zongjun Gao and Jiutan Liu

Received: 17 November 2024
Revised: 12 December 2024
Accepted: 19 December 2024
Published: 23 December 2024

1. Introduction

Slovenia is home to a remarkable diversity of subterranean species known as troglobionts, making it a hotspot for subterranean biodiversity [1,2]. These subterranean species often have very restricted habitats, frequently existing in only small areas or even a single location [3]. One notable species is the olm or proteus (*Proteus anguinus* Laurenti), an endemic amphibian that inhabits the subterranean water environments of the Dinaric Karst. The white olm (*Proteus anguinus anguinus* Laurenti) is commonly found throughout this region of the Western Balkans. The black olm (*Proteus anguinus parkelj* Sket & Arntzen) was discovered in 1986 in the karst spring of the Dobličica River, nearly three hundred years after the white species was first documented. The habitat of the black olm is confined to a small area (4 km^2) of karst springs in the Bela Krajina region in southern Slovenia, making it particularly vulnerable to environmental changes and human activities [4–6].

The black olm is characterised by its darker pigmentation and thrives in stable, oxygen-rich, and unpolluted water environments with temperatures ranging from 8 °C to 11 °C [7]. According to national legislation on protected wildlife, white and black olms are legally recognised as protected and vulnerable species in Slovenia. Due to their rarity, they are classified as vulnerable on the Red List of the Republic of Slovenia. The primary threat to their populations comes from high concentrations of nitrates originating from agricultural runoff and urban areas with poorly managed wastewater disposal and illegal waste dumps [6,7]. Elevated nitrate levels can lead to eutrophication, which causes a reduction in dissolved oxygen in the water, risking suffocation for the olms [8–10]. Notably, the acceptable limit for nitrate concentration in water for olm ecosystems is 9.2 mg NO_3^-/L, five times lower than the limit for drinking water at 50 mg NO_3^-/L [11].

The selection of black olm as the focal species of this study was driven by several ecological and conservation reasons, including (i) biodiversity maintenance contributing to overall ecosystem health and stability, (ii) indicator species providing valuable information about the state of the ecosystem, and (iii) cultural and ethical values, helping to preserve cultural heritage and promote ethical stewardship of the environment.

Karst is a landscape formed by the dissolution of limestone and dolomite by slightly acidic water, leading to features like lakes, caves, and sinkholes [12,13]. About 43% of the land in Slovenia is karst, mainly in the southwest, with Alpine, Dinaric, and Transitional karst types. The soil in karst areas varies, featuring undeveloped types like rendzinas in forests and jerovica or terra rossa in vineyards. Lower karst areas have more developed eutric brown soils [13]. Karst soils are generally shallow, diverse, rich in organic matter, and slightly acidic, but they often experience water shortages during droughts [14].

Karst springs connect to surface waters through subterranean channels. These systems are crucial for drinking water, supporting 20–25% of the world's population. They consist of interconnected areas with different shapes and water flow [15]. Water enters karst aquifers slowly through cracks or quickly through sinkholes, making them vulnerable to pollution but vital for ecological balance [6,16]. Pollutants from septic tanks and agricultural land can contaminate karst groundwater directly or via surface water [17–20]. Bellow the soil profile, the gravitational water flow transports pollutants into the aquifer, which reappear at the karst spring [19]. Fertilisers can leach into the groundwater and decrease water quality due to poor waste management practices and agricultural methods [21–23].

To safeguard groundwater, which constitutes a vital source of drinking water in Slovenia (exceeding 95%), the entire country is designated as a vulnerable nitrate area [24]. Following the Nitrates Directive (91/676/EEC) [25], aimed at protecting water from nitrate contamination from agricultural activities, all farms must adhere to specific regulations. These regulations mandate that fertilisers be applied only during periods when crops require nutrients for growth, that fertilisation rates be adjusted to align with the nutritional needs of the plants, that appropriate machinery be employed, and that sufficient storage capacities for livestock manure be maintained during the non-growing season when fertilisation of agricultural land is prohibited [26].

Current research on terrestrial subterranean karst systems primarily focuses on several key areas: defining hydrogeological characteristics [6,12,16]; identifying drivers of species richness [1,2]; assessing groundwater quality by measuring nitrogen concentrations and other contaminants, as well as determining pollution sources [7,15,18,20–23]; examining the toxicity of nitrogen to common organisms [9]; and using SWAT modelling to identify critical source areas and address non-point source pollution [19]. One of the most comprehensive works on agriculture and karst systems is the book *Karst Management*, published in 2011 [17], which discusses various agricultural activities that may impact soil and water in karst regions. This study advances our understanding of how implementing appropriate agricultural practices can protect the vulnerable karst groundwater habitat of the black olm from nitrate pollution. We investigate the potential nitrate concentrations in karst springs that are vital habitats for the black olm by implementing a range of alternative scenarios. We can better protect this unique species and its environment by understanding these dynamics.

This research seeks to identify the hotspots of nitrate leaching within the vulnerable groundwater habitat of the black olm residing in the Dobličica River aquifer. Additionally, it evaluates nitrate leaching by modelling various scenarios that incorporate adaptations in agricultural crop rotations and alterations in land use. Ultimately, the study aims to establish agricultural measures that will effectively reduce nitrate leaching and enhance the protection and improvement of the groundwater habitat of the black olm.

2. Materials and Methods

2.1. Study Area

The study area is located in south-eastern Slovenia (74 km^2), west of Črnomelj, within the Dobličica River Basin, which is part of the larger Lahinja River Basin. The altitude ranges from 140 to 1051 m, featuring several karst springs fed by the deep karst of Poljanska Gora (Figure 1a). Key springs include, from north to south, Otovski breg (157 m.a.s.l.), Pački breg (159 m.a.s.l.), Jelševniščica (142.5 m.a.s.l.), Obršec (145.1 m.a.s.l.), and Dobličica (141 m.a.s.l.), which is the primary drinking water source for the Bela Krajina region.

The terrain is structured by typical Dinaric folds and faults that spread in the northwest–southeast direction, so the movement of water through the system is relatively fast and can transfer pollutants from the surface to the aquifer in a short time [6]. Geologically, the area is characterised by Mesozoic-age carbonate rocks, which comprise the shallow marine carbonate platform. The climate of Bela Krajina is moderate continental, with average annual temperatures around 10.5 °C (July 21.2 °C, January −0.2 °C) and average annual rainfall of 1285 mm, peaking in spring and autumn.

The soil in the region is often shallow and stony, restricting its suitability for intensive agriculture (Figure 1d). The soil in the region formed on limestone and dolomite is often shallow and stony, with leached soils (Luvisol) covering 60% of the area (forest), while brown carbonate soil (Cambisol) makes up 30%, primarily in mixed forest and agricultural settings. The remaining 14% of the area includes district brown soils on Pliocene deposits (7%), rendzina (5%), and alluvial soils (2%). Land use is dominated by forest (81%), followed by meadows (8%), fields (3%), urban areas (2.5%), extensive orchards (2%), vineyards (1%), and other (3.5%) (Figure 1c). In dolinas and sinkholes, various crops are cultivated. Forest dominates steeper terrain and higher altitudes. Typical crop rotations consist of maise, cereals, and clover–grass mixtures for livestock farming. Winter wheat, silage maise, and winter barley comprise about 80% of cropland, with clovers, vegetables, and potatoes covering the rest [27]. The livestock density is 0.48 livestock units (LU) per hectare, including cattle and sheep.

2.2. SWAT Modelling

The SWAT 2012 (Soil and Water Assessment Tool) program, used in this study, is a robust model designed to predict hydrological processes in natural and human-influenced basins [28]. It integrates data from climate prediction models, soil characteristics, and land use scenarios to assess their effects on hydrology [29]. Developed by the USDA Agricultural Research Service, SWAT is widely applied for soil erosion control, non-point source pollution, and regional watershed management [30]. Its relevance has increased in the European Union since the 2000 Water Framework Directive, given its adaptability to various regions and environmental conditions [31,32].

The model's basic unit is the hydrological response unit (HRU), defined by consistent soil properties, land use, and slope. The number of HRUs varies based on the combination of these factors. SWAT simulates precipitation, runoff, infiltration, soil water content, evapotranspiration, and crop growth. It also models the transport of sediments, nutrients, and pesticides from land to water through surface runoff and groundwater flow [33]. Its flexibility allows for application in various basin sizes and land management practices [19].

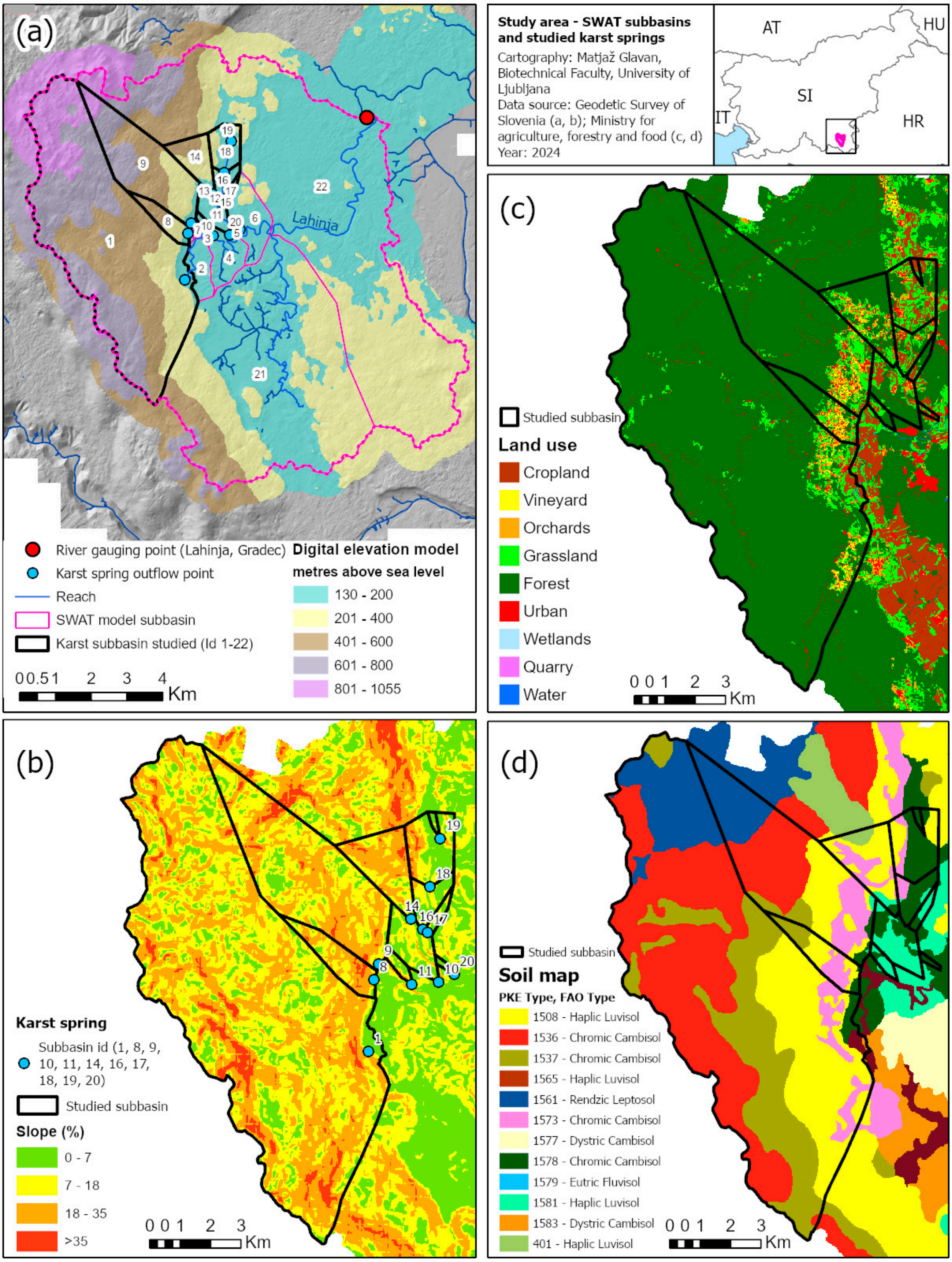


Figure 1. Geographic characteristics of study area presenting (**a**) topography, river network, SWAT model subbasin numbers (1–22), main river gauging point and karst springs outflow points, (**b**) slope and karst springs (with SWAT model subbasin id number), (**c**) land use, and (**d**) soil type.

Setting up the SWAT model entails several crucial steps to ensure its accuracy and reliability in simulating hydrological and nutrient cycling processes [28,30]. First, data collection is essential; this involves gathering observed data for the variables to be simulated, such as streamflow or nutrient concentrations, as this information is vital for calibration and validation. Next, the model configuration requires inputting the necessary data to reflect the studied river basin's physical characteristics accurately. The calibration phase adjusts model parameters to align simulated outputs with observed data, which can be accomplished manually or by utilising automated calibration tools like SWAT-CUP. This phase includes sensitivity analysis to pinpoint parameters that significantly affect model outputs, iterative adjustments of these sensitive parameters to enhance the fit between simulated and observed data, and the utilisation of performance metrics such as the coefficient of determination (R^2), Nash–Sutcliffe Efficiency (*NSE*), and Percent Bias (*PBIAS*) to assess the model's efficacy during calibration. Following calibration, validation is conducted to test the model with a distinct set of observed data not used in the calibration process, ensuring accurate simulations for conditions outside this period. This step comprises running the model using calibrated parameters for the validation period, comparing the simulated outputs against the observed data and employing statistical measures (R^2, *NSE*, *PBIAS*) to evaluate the model's performance and robustness, confirming consistent results across both calibration and validation periods. Adhering to these steps guarantees that the SWAT model is thoroughly calibrated and validated, yielding credible simulations for hydrological studies [28].

2.3. Model Input Database

The SWAT model requires a comprehensive dataset due to the complexities of simulating hydrological processes. The digital elevation model (DEM) serves as the basis for defining the basin area (Table 1). In this karst region, we utilised subbasin areas of karst springs developed by hydrogeologists from the Geological Institute of Slovenia and karstologists from the ZRC SAZU Institute for Karst Research (Figure 1).

We adjusted the parameters regulating water distribution among surface flow, subsurface lateral flow, and groundwater to accurately simulate water movement in karst systems. The official river network from the Water Directorate of the Republic of Slovenia was employed to map riverbeds and spring locations. Soil and land use data were obtained from the Ministry of Agriculture, Forestry, and Food. We also included local agricultural rotations and practices, such as ploughing and fertilisation, obtained from the regional Agricultural Advisory Service Novo mesto. River flow data and nitrate measurements were acquired from the Slovenian Environmental Agency and ZRC SAZU Karst Research Institute (Table 1). Data regarding nitrate concentration in water within the studied river basin were limited. The only reliable, accessible, and trusted measurements were conducted in 2021 and 2022.

Weather data were collected from the Črnomelj-Dobliče meteorological station, with solar radiation data sourced from the nearest station in Novo mesto.

Soil data in the SWAT model comprises physical and chemical properties. Physical properties influence water and air movement within the soil profile, affecting the hydrological cycle in hydrological response units (HRUs), while chemical properties establish initial concentrations of nitrogen (N), phosphorus (P), agronomic chemicals, and heavy metals. Although the national soil map provided initial input data, we utilised the pedotransfer function [34,35] to estimate additional missing physical properties of the soil, including bulk density, water retention, and hydraulic conductivity.

2.4. Model Setup, Calibration, and Validation

For modelling, we employed ArcSWAT version 2012.10.8.26, an ArcGIS extension and interface specifically designed for the SWAT model [28]. The reliability of the SWAT model was enhanced through a calibration process, as numerous model parameters could not be measured directly due to high equipment costs and a lack of available human resources.

The calibration was conducted utilising observed data and subsequently validated with an independent dataset. The model is considered appropriate if the observed and simulated data discrepancies are minimal.

Table 1. Input data for setting up a model in the SWAT program.

Weather	**Unit**	**Data Source**
precipitation	mm day^{-1}	Slovenian Environmental Agency (ARSO) Gauging stations Dobliče (precipitation, temperature, wind, relative humidity), Novo mesto (sun)
min. and max. temperature	°C day^{-1}	
relative humidity	fraction	
global radiation energy (sun hours)	(h day^{-1})	
average wind speed	km day^{-1}	
Soil		
digital soil map, soil horizons, colour	.shp	Ministry of Agriculture, Forestry and Food of the Republic of Slovenia (MKGP)
soil depth, roth depth	cm	
texture (clay, silt, sand), organic matter	mass %	
soil density	cm^3 cm^{-3}	Calculation of pedotransfer functions based on digital soil map data (MKGP)
field capacity, wilting point	cm cm^{-1}	
saturated hydraulic conductivity	cm h^{-1}	
soil erosivity—MUSLE		calculation
Crop production techniques		
crop type and rotation, sowing/planting/harvesting time		Farmers Agricultural advisory service (KGZ Novo mesto)
fertiliser type and method of application, equipment use scheduler, depth of cultivation		
Land		
actual land use	.shp	MKGP
digital elevation model	raster	Geodetic Survey RS (GURS)
Water		
river flow—Lahinja gauging station Gradac (1992–2022)	m^3/s	ARSO
karst springs (2021–2022)		Institute for Karst Research (ZRC SAZU)
nitrate content—Lahinja gauging station Gradac (2021–2022)	(mg/L NO_3^-)	ARSO
monthly nitrate content (09/2021–11/2022)		ZRC SAZU
karst springs subbasins	.shp	ZRC SAZU

Despite its ecological significance, the availability of measured data for the study area is limited. Average daily flow data were obtained from a single gauging station on the Lahinja River (Gradac) managed by the Slovenian Environmental Agency, located downstream of the tributary Dobličica River, serving as a base model development reference. Nitrogen content data were available only for the year 2022. The model operated with a 30-year dataset from 1 January 1992 to 31 December 2022, divided into warm-up (1993–1997), calibration (1998–2010), and validation (2011–2022) phases. For the 11 karst springs observed (subbasins), average annual flow data were obtained from the Institute for Karst Research. They describe data as derived from water balance calculations for 2021–2022, and monthly nitrate data were collected from October 2021 to November

2022. Calibration was performed over the entire 2021–2022 dataset. Due to a lack of data, validation could not be performed.

The initial calibration was conducted manually to evaluate the effects of SWAT parameters on simulation results, followed by automatic calibration using SWAT-CUP Premium and then manual adjustments for accuracy. The SPE (SWAT Parameter Estimator) algorithm facilitated sensitivity analysis, calibration, and validation [36].

The base model for the Lahinja River Basin includes 22 subbasins and 1125 Hydrological Response Units (HRUs). Our analysis focused on 11 subbasins within the Dobličica River area (Figure 1). This focus was necessitated by the fact that only the hinterland of the selected subbasins (1, 8, 9, 10, 11, 14, 16, 17, 18, 19, 20) has a significant impact on the habitat of the black olm.

2.5. Scenarios

Based on a comprehensive literature review and analyses of data from prior studies concerning the black olm, supplemented by a field visit, scenarios were developed employing the SWAT model to evaluate the effects of crop rotations and changes in agricultural land use on nutrient leaching and groundwater quality at both the basin and subbasin levels [27]. A total of 11 alternative scenarios were developed (Table 2). The crop rotation change scenarios (R) focus on adapting agricultural technologies, expanding crop rotations, and managing fertilisation and tillage on arable land (scenarios 1–5). The agricultural land use change scenarios (E) encompass a broader range of measures that involve shifts in land use (such as converting cropland to grassland or forest), which subsequently affect cultivation intensity (scenarios 6–11). This scenario analysis thoroughly examines the impact of various potential measures on the amount of nitrogen leached from soils into groundwater and surface waters, thereby influencing their quality and the ecosystem of the black olm habitat.

When selecting the crop rotation change scenarios, the environmental effects of reducing nitrogen losses to the environment (effectiveness) were primarily followed, followed in second place by the preservation of the amount of biomass needed as feed for animal nutrition, and in third place, the ease of introduction into agricultural practice. Only environmental objectives were primarily followed when selecting the agricultural land use change scenarios. The exception is scenario E6, where the change sought to economically improve the situation of farms that were prepared to change extensive agricultural land (fields into meadows, meadows into forests) in hot spots that are shallow, steep, and at higher altitudes. Adverse economic effects on agricultural holdings (costs) that could arise from introducing measures would be mitigated by direct payments within the agricultural or environmental policy framework, providing a safety net against potential risks.

This research employed the SWAT model to simulate the nitrogen cycle in agricultural soil for each alternative scenario, thereby facilitating insights into the transport of nitrates within the soil and subbasins. The simulation results encompass hydrological processes, including surface runoff, lateral runoff, percolation, and nitrogen cycle processes, such as plant uptake, nitrification, denitrification, and nitrate leaching. The model effectively quantifies the movement of water and nitrogen below the root zone, where it becomes unavailable to plants, ultimately entering groundwater flow and, subsequently, surface waters. The data collection process for smaller, spatially defined hydrological response units (HRUs) has enabled the identification of critical nitrogen leaching points.

The results derived from the various scenarios were thoroughly analysed and presented in tabular and graphical formats. A map for the entire study area was generated, indicating regions where nutrient leaching is anticipated, given the constraints of the input data, along with management recommendations for critical areas.

Table 2. Scenarios of agricultural crop rotations and agricultural land use change.

Scenarios			
No.	**Name**	**Description**	
		Arable Land	**Grassland**
0	BASE (B)	4-year rotation, no greening in 2nd year after winter wheat maize/wheat + no greening/maize/barley + clover–grass mix	3 cuts
AGRICULTURAL CROP ROTATION CHANGE SCENARIOS			
1	ROTATION (R1)	2-year rotation maise/winter barley + summer maize + clover–grass mix (CGM)	3 cuts
2	ROTATION 1 (R2)	B + CGM in 2nd year (maize/wheat + CGM/maize/barley + CGM)	3 cuts
3	ROTATION 2 (R3)	R2 + 20% decrease in fertilisation (maize/wheat + CGM/maize/barley + CGM)	3 cuts
4	ROTATION 3 (R4)	6-year rotation R3 + additional 2 years of CGM (maize/wheat + CGM/maize/barley + CGM/CGM/CGM	3 cuts
5	ROTATION 4 (R5)	6-year rotation R4 + winter fodder peas (WFP) replaced winter wheat (maize/WFP + CGM/maize/barley + CGM/CGM/CGM	3 cuts
AGRICULTURAL LAND USE CHANGE SCENARIOS			
6	EXTENSIVE 1 (E1)	B + selected fields into grassland (3 cuts) (slope > 7%, Soil PKE type: 1508, 1536, 1537, 1561, 1573)	3 cuts
7	EXTENSIVE 2 (E2)	B + all fields into grassland (3 cuts)	3 cuts
8	EXTENSIVE 3 (E3)	B + selected fields into unfertilised grassland (1 cut) (slope > 7%, Soil PKE type: 1508, 1536, 1537, 1561, 1573)	1 cut
9	EXTENSIVE 4 (E4)	B + all fields into unfertilised grassland (1 cut)	1 cut
10	EXTENSIVE 5 (E5)	B + E4 + all grassland into forest	/
11	EXTENSIVE 6 (E6)	R5 + selected grasslands into fields (slope < 7, Soil PKE type: 1578, 1579, 1583)	3 cuts

The evaluation of the scenario results was based on the outputs of the SWAT model, employing the following indicators:

− Nitrate nitrogen transported into the main channel in the groundwater loading from the HRU (kg N/ha per year (NO3GW);
− Nitrate nitrogen leached from the soil profile (kg N/ha). Nitrate that leaches past the bottom of the soil profile during the time (NO3L);
− Average annual total biomass (dry matter) per HRU (metric tons/ha) (BIOM);
− Total nitrogen (TOT_N) transported with surface water flow from the subbasin (kg N/year);
− Nitrate nitrogen (NO3_OUT) transported by surface water flow from the subbasin (kg N/year).

2.6. Model Performance Objective Functions

To assess the model predictions for streamflow, we utilised three statistical methods: the coefficient of determination (R^2), the Nash–Sutcliffe efficiency (*NSE*), and percent bias (*PBIAS*). These methods are well-established for evaluating the performance of hydrological models [37].

The Coefficient of Determination (R^2) measures the proportion of the variance in the observed data that is predictable from the model. It ranges from 0 to 1, where a value closer to 1 indicates a better fit. An R^2 value greater than 0.60 is generally considered satisfactory for hydrological models, indicating that the model explains at least 60% of the variance in the observed data. The equation for R^2 is

$$R^2 = 1 - \left(\frac{\sum_{i=1}^{n} (O_i - P_i)^2}{\sum_{i=1}^{n} (O_i - \overline{O})^2} \right) \quad (1)$$

where (Oi) is the observed value, (Pi) is the predicted value, and $\overline{O}$ is the mean of the observed values.

Nash–Sutcliffe Efficiency (*NSE*) is a normalised statistic that determines the relative magnitude of the residual variance compared to the measured data variance. It ranges from $-\infty$ to 1, where 1 indicates a perfect match between the model and the observations. An *NSE* value greater than 0.50 is deemed satisfactory, suggesting that the model predictions are more accurate than the mean of the observed data. The equation for *NSE* is

$$NSE = 1 - \frac{\sum_{i=1}^{n} (O_i - O_i)^2}{\sum_{i=1}^{n} (O_i - \overline{O})^2} \quad (2)$$

Percent Bias (*PBIAS*) measures the average tendency of the simulated data to be larger or smaller than their observed counterparts. It is expressed as a percentage. Positive values indicate model overestimation bias, while negative values indicate underestimation bias. The equation for *PBIAS* is

$$PBIAS = 100 \times \frac{\sum_{i=1}^{n} (P_i - O_i)}{\sum_{i=1}^{n} O_i} \quad (3)$$

Moriasi et al. [37] established the following performance ratings for each statistical measure. Generally, a model's simulation is considered satisfactory if the following criteria are met: $R^2 > 0.60$, $NSE > 0.50$, *PBIAS* within ±15% for streamflow, and *PBIAS* within ±30% for nitrogen predictions [38].

3. Results and Discussion

3.1. Sensitivity Analysis, Calibration, and Validation

In the sensitivity analysis process, greater emphasis was placed on parameters contributing to the results' variability during the calibration phase (Table 3). The performance indicators for the model calibration regarding the flow and nitrate nitrogen levels at the Lahinja River gauging station and karst springs are presented in Table 4. For the Lahinja River, daily measured data from the Gradac gauging station were used. The model calibration from 1998 to 2010 yielded the following results: *NSE* 0.59, R^2 0.61, and *PBIAS* 0.09. During the validation period from 2011 to 2022, these values improved to *NSE* 0.69, R^2 0.71, and *PBIAS* −2.28 (Table 4, Figure 2). A negative *PBIAS* during validation indicates that the model slightly overestimates the measured flow values. Overall, these results demonstrate a good agreement between the model and the measured data.

The annual average flows of karst springs from 2020 to 2022 exhibited differences between the measured and simulated values, allowing for an evaluation of flow excess or deficiency (Table 4). Except for three springs, all other values fell within the satisfactory range for *PBIAS* (±25%) [37]. It is important to note that the average annual flow values for karst springs were calculated based on the water balance between precipitation and evapotranspiration (ET), with occasional adjustments from flow measurements, which were then compared to the simulated flows. Regular flow measurements at karst springs should be conducted in the future to mitigate uncertainty. Additionally, the heterogeneity of subterranean water flows and the mixed overflows at varying water levels contribute to further uncertainty in flow assessments, a phenomenon that has also been observed by karstologists and hydrologists [39,40].

Table 3. SWAT parameters, range (min. and max. value), default and final values used in the model for flow calibration and validation and nitrogen calibration at Gradac gauging station.

SWAT File	SWAT Parameters		Range	Default Value	Final Value
		River Flow			
.gw	GW_DELAY	Groundwater delay	0–500	31	3
	ALPHA_BF	Baseflow alpha factor	0–1	0.048	0.70
	GWQMN	Threshold depth of water in the shallow aquifer required for return flow to occur	0–5000	1000	500
	GW_REVAP	Groundwater "revap" coefficient	0.02–0.2	0.02	0.02
	RCHRG_DP	Deep aquifer percolation fraction	0–1	0.05	0.01
	REVAPMN	Threshold depth of water in the shallow aquifer for "revap" to occur	0–1000	750	750
.mgt	CN2	SCS runoff curve number for moisture condition 2	0–100	variable	−14.4%
.hoe	ESCO	Soil evaporation compensation factor	0–1	0.95	1.00
	SURLAG	Surface runoff lag time	0.01–24	4	4.00
.bsn	SFTMP	Snowfall temperature (°C)	–5–5	1	2.3
	SMTMP	Snow melt base temperature (°C)	–5–5	0.5	3.515
	SMFMX	Maximum melt rate for snow during the year (occurs on summer solstice) (°C)	0–10	4.5	2.377
	SMFMN	Minimum melt rate for snow during the year (occurs on winter solstice) (°C)	0–10	4.5	3.457
	TIMP	Snowpack temperature lag factor	0–1	1	0.203
	SNOCOVMX	Minimum snow water content that corresponds to 100% snow cover	0–500	1	31.429
		Nitrogen load			
.bsn	CMN	Rate factor for humus mineralisation of active organic nitrogen	0.001–0.003	0.0003	0.003
	RCN	The concentration of nitrogen in rainfall	0–15	0.9	1.150
	CDN	Denitrification exponential rate coefficient	0–3	0.0003	1.4
.gw	HLIFE_NGW	Half-life of Nitrate in the shallow aquifer [days]	1–365	0	32.5–365
.sep	ISEP_TYP	The type of septic system	1–100	1	1
	SEP_DEN	Number of septic systems per square kilometre (only urban land use)	0.001–500	1.5	280
	ISP_OPT	Current condition of OWS (1 = active septic, 2 = failing septic, 0 = non-septic)	0–2	0	1
	SEP_CAP	Number of permanent residents in the house	1–10,000	2.5	2.5
Databases SepticWQ	SPTQ	Septic tank effluent (STE) flow rate (m^3/capita/day)	0–1	0.227	0.227
	IDSPTTYPE	Type of a septic system	1–3	1	1

Table 4. Performance indicators of calibration for measured river flow at Gradac gauging station (m^3/s) and average annual flow of karst springs and comparison of nitrate nitrogen load (kg N/day) at karst springs.

Objective Functions of Model Performance Observed vs. Simulated			
	NSE	R^2	*PBIAS*
Gradac gauging station—River Lahinja—daily data			
Flow Calibration (1998–2010)	0.59	0.61	0.09
Flow Validation (2011–2022)	0.69	0.71	−2.28
Nitrate nitrogen Calibration (2021–2022)	-	-	12.74
Karst springs—Calibration (2021–2022)—average annual values			
		PBIAS	
Subbasin	**Name**	**River flow**	**Nitrate nitrogen**
1	Dobličica	7.56	0.01
8	Obršec	−5.60	0.04
9	Jelševniščica	3.14	−0.15
11	Janževe loke	0.69	−0.28
10	Šprajcarjev zdenec	27.86	5.66
14	Talački breg	1.02	−0.04
16	Pački breg	−14.25	−0.08
18	Otovski breg	−11.01	−0.12
17	Brežiček	−38.88	0.07
19	Stobe	−80.54	−0.20
20	Planinc	−57.69	−0.16

The calibration results for the daily transfer of nitrate nitrogen by water flow from 2021 to 2022 demonstrate a strong agreement, confirming the reliability of the model predictions (Table 4, Figure 3). Nitrate nitrogen levels in water are observed to increase during periods of high flow, a phenomenon associated with elevated precipitation that results in enhanced nitrogen leaching from agricultural soils into the aquifer (Figure 3a). In contrast, the nitrate nitrogen concentrations within the Jelševnik karst spring are characterised by low and stable values, indicating that the natural background is the primary source of nitrogen (Figure 3b). While the model does a good job of predicting nitrate nitrogen dynamics, some local variability may exist that it does not fully capture. The Percent Bias (*PBIAS*) values for the Gradac gauging station and karst springs are 12.74 and less than 6, respectively (Figure 2). This consistency between the measured and simulated values confirms the model's accuracy and usefulness in simulating nitrate nitrogen levels. Such a high level of accuracy is essential for effective water quality management in the studied karst area, as controlling nitrate concentrations is crucial for protecting both the environment and human health. It is crucial to acknowledge that simulated values were compared against observed data using monthly data from September 2021 to November 2022 (Table 1). The parameter HLIFE_NGW, which represents the half-life of nitrate nitrogen in a shallow aquifer, had the most significant impact on achieving this strong agreement (Table 3). This is in line with studies that provide insight into the response and transport of nitrate in karst groundwater to rainfall events and denitrification [41], as well as on karst hydrological and biogeochemical time lag impacts resulting in time-delayed impact of measures (rotation, land use change) on water quality [21].

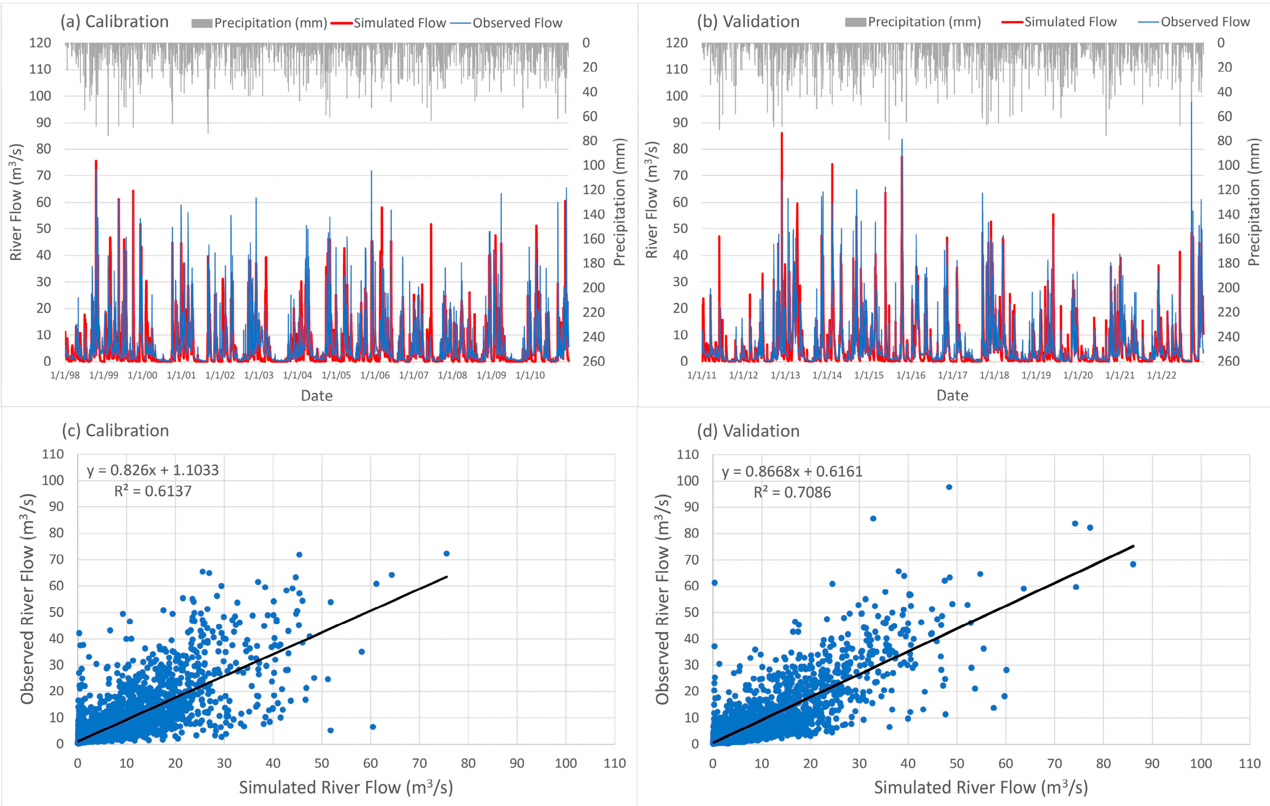


Figure 2. Display of measured and simulated flow data at the Gradac gauging station for the period of (**a**,**b**) calibration (1998–2010) and (**c**,**d**) validation (2011–2022) on a daily time step.

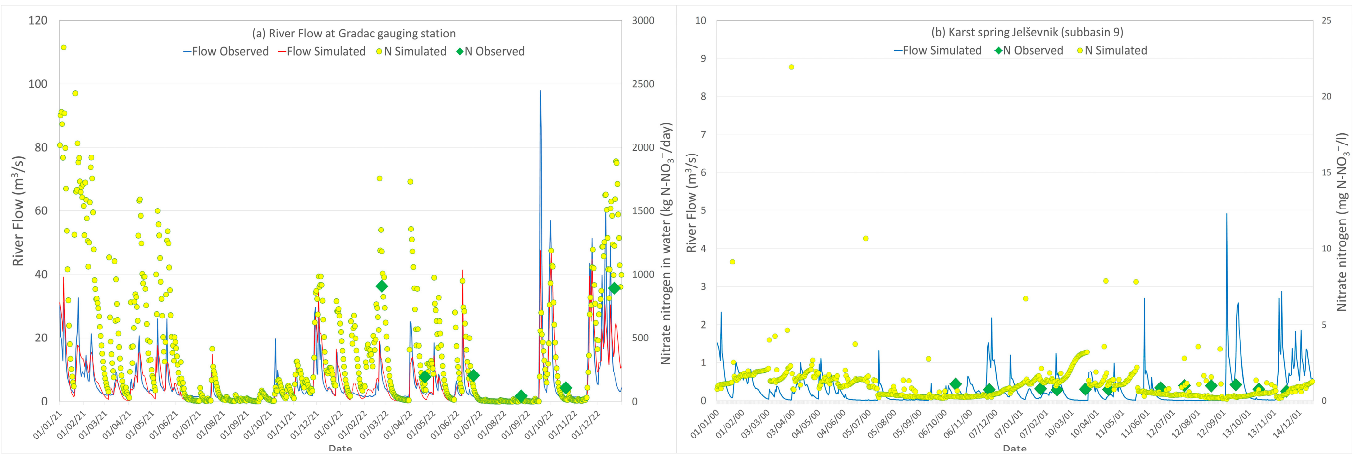


Figure 3. Comparison of observed and simulated data on (**a**) the nitrate nitrogen yield (kg N-NO_3^-/day) and water flow at the Gradac measuring station (subbasin 22) and (**b**) the nitrate nitrogen concentration (mg N-NO_3^-/day) and water flow at the Jelševnik karst spring (subbasin 9) on a daily time step.

3.2. Assessment of Agri-Environmental and Land Use Scenarios

3.2.1. Nitrate Leaching from the Hydrological Response Unit into Groundwater Flow

The results indicate that implementing alternative crop rotation scenarios (R2 to R5) can effectively reduce the leaching of nitrate nitrogen into groundwater. The most significant reductions are observed in scenarios R3 and R4. Detailed effects of the different scenarios are presented in Table 5.

Table 5. Comparison of results between baseline and alternative scenarios of agricultural crop rotation change for nitrate transported into the main channel in the groundwater loading from the HRU (NO3GW) (kg N/ha year), nitrate leached from the soil profile (NO3L) (kg N/ha year), and total plant biomass (BIOM) (metric tons/ha year).

Simulated Annual Averages (1998–2022)	**CROPLAND**										
	Subbasin Name and Number										
	Dobličica	**Obršec**	**Jelševnik**	**Janževe Loke**	**Šprajcarjev Zdenec**	**Talački Breg**	**Pački Breg**	**Brežiček**	**Otovski Breg**	**Stobe**	**Planinc**
	1	**8**	**9**	**10**	**11**	**14**	**16**	**17**	**18**	**19**	**20**
Nitrate transported into the main channel in the groundwater loading from the HRU (NO3GW) (kg N/ha year)											
BASE model (kg N/ha year)	23	21	15	8	32	25	7	18	16	8	22
StDv	17	16	12	7	21	18	7	14	13	7	15
SCENARIOS	Change (%) in the amount of nitrate nitrogen (N-NO_3^-)										
R1	67	64	63	70	68	69	70	66	66	69	62
R2	−16	−15	−12	−18	−16	−17	−17	−18	−18	−17	−16
R3	−38	−38	−35	−39	−37	−39	−40	−39	−40	−36	−36
R4	−68	−68	−66	−67	−66	−68	−68	−67	−68	−65	−66
R5	−52	−52	−50	−50	−51	−52	−52	−51	−52	−48	−49
Nitrate leached from the soil profile (NO3L) (kg N/ha year)											
BASE model (kg N/ha year)	61	65	73	66	64	60	66	68	61	72	75
StDv	46	48	58	45	44	44	47	46	45	46	47
SCENARIOS	Change (%) in the amount of nitrate nitrogen (N-NO_3^-)										
R1	70	68	64	69	70	72	70	70	71	66	64
R2	−13	−12	−8	−13	−14	−15	−14	−13	−14	−12	−11
R3	−35	−35	−31	−34	−35	−37	−36	−34	−36	−32	−31
R4	−66	−65	−62	−64	−64	−66	−66	−64	−65	−62	−62
R5	−50	−49	−47	−48	−48	−50	−50	−48	−49	−46	−46
Total plant biomass (BIOM) (metric tons/ha year)											
BASE model (kg N/ha year)	25	25	24	26	26	26	26	26	26	26	26
StDv	13	13	13	13	13	13	13	13	13	13	13
SCENARIOS	Change (%) in the amount of biomass (dry matter)										
R1	16	16	16	17	17	17	17	17	16	16	16
R2	18	18	18	19	19	18	18	19	18	18	18
R3	14	14	15	15	15	15	15	15	14	14	14
R4	−5	−4	−3	−4	−4	−4	−4	−4	−5	−5	−4
R5	−1	0	1	−1	−1	−1	0	−1	−1	−1	−1

Scenario descriptions and abbreviations are explained in Table 2; red—increase in N leaching, blue—decrease in N leaching; green—increase in biomass, yellow—decrease in biomass.

The base scenario (B) represents the current situation without additional measures to reduce nitrate nitrogen leaching. The highest levels of nitrate nitrogen (kg N/ha per year) detected in groundwater transported from HRUs into surface waters (NO3GW) were observed in subbasins 1, 11, and 14 (Table 5).

Scenario R1 introduces a two-year corn crop rotation, which leads to more intensive fertiliser use and a significant increase in the amount of leached nitrate nitrogen across all subbasins. For instance, in subbasin 1, NO3GW rises by 67%, from approximately 23 kg/ha per year to about 40 kg/ha per year. The amount of nitrate leaching (NO3L) increases by 70%. Additionally, the average annual dry-weight biomass (BIOM) production increases by up to 17%.

Scenario R2 incorporates winter greening practices. This scenario reduces the amount of nitrate nitrogen in groundwater by 12% to 18%, while NO3L declines by 8% to 15%. With winter greening, the average annual biomass production increases by up to 19%.

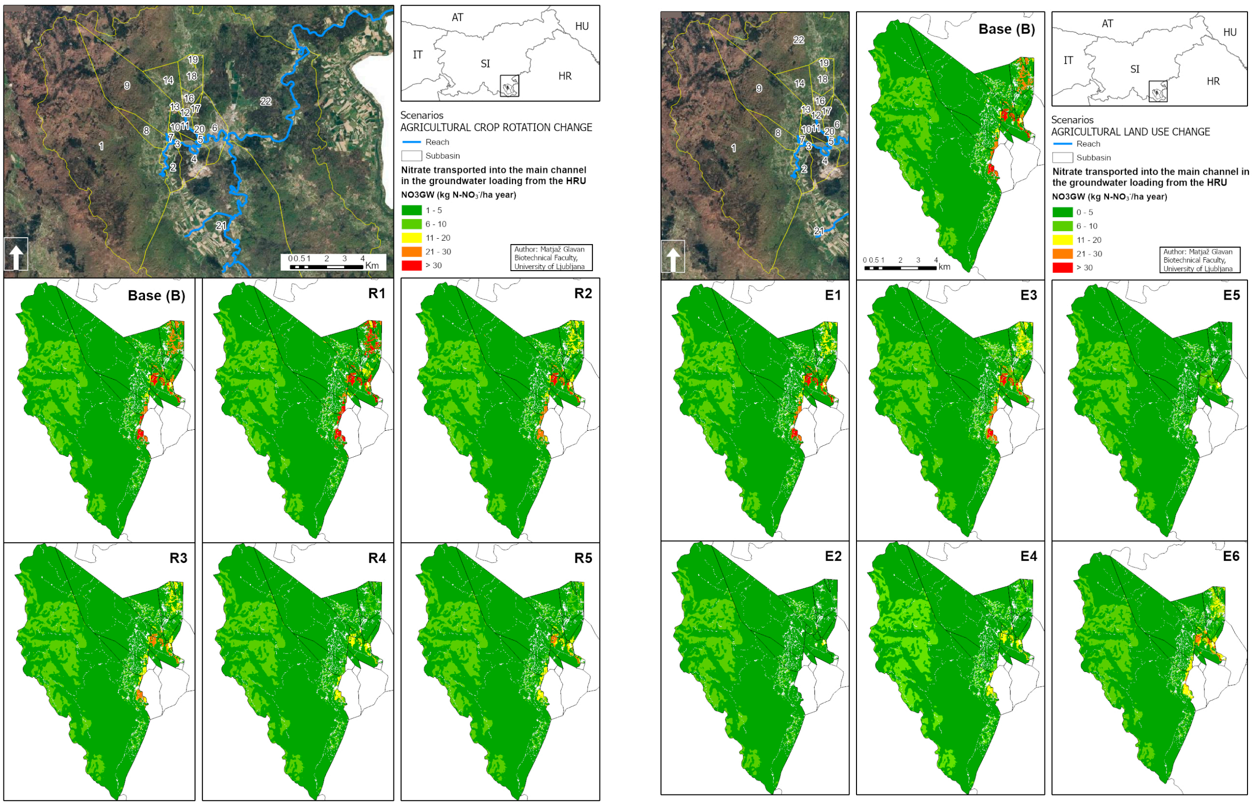
Scenario R3 combines winter greening with a 20% reduction in fertiliser use. In this scenario, NO3GW decreases by 35% to 40%, and NO3L is reduced by 31% to 37%. The average annual biomass production increases by up to 15%.

Scenario R4 extends the CGM growing period by an additional two years. This scenario results in the highest overall reduction in nitrate nitrogen, with NO3GW decreasing by

65% to 68% and NO3L reduced by 62% to 66%. The average annual biomass production decreases by 5%, primarily due to less frequent sowing of maise.

Scenario R5 replaces winter wheat with winter fodder peas. In this case, NO3GW decreases by 48% to 52%, and NO3L is reduced by 46% to 50%. However, the average annual biomass production declines by up to 1%, as pea yield is lower than wheat.

Figure 4 illustrates the areas critical for nitrogen leaching from the soil. The green shades represent hydrological response units (HRUs) with lower leaching potential and, consequently, lower levels of nitrate nitrogen in groundwater. In contrast, the red shades highlight HRUs with high nitrogen leaching potential, requiring immediate attention and implementing better agricultural practices for fertiliser use. These critical areas can be identified as hotspots where more focused management and advice are necessary. Alternative scenarios, particularly R2, R3, and R4, show an increase in green areas, indicating lower nitrate nitrogen leaching compared to the baseline scenario characterised by intensive fertilisation. The measures proposed in scenarios R2 and R3 further demonstrate the effectiveness of these strategies in improving water quality and reducing the environmental impact of agriculture while also supporting enhanced biomass production.



(**a**) (**b**)

Figure 4. Comparison of the basic scenario with alternative scenarios of (**a**) agricultural crop rotation change (R) and (**b**) agricultural land use change (E) in relation to the average nitrate nitrogen transported (kg N-NO_3^-/ha per year) from HRUs in the groundwater flow to the surface water flow in the study area of 11 subbasins.

Key factors influencing the rate of nitrate leaching and the soil's capacity for plant growth include soil depth, hydrological group, and soil texture (Table 6). Soils with greater water and nutrient retention capacity, such as deep loamy soils, yield higher biomass while simultaneously reducing nitrate nitrogen leaching [42,43]. This information is crucial for planning and implementing sustainable agricultural practices in a spatially considerate manner.

Table 6. The impact of soil properties in relation to land use (cropland grassland) on nitrogen leaching below the soil profile and the plant biomass yield for the base scenario (B).

Soil Type	Soil Properties						Nitrate Leached Below Soil Profile (NO3L)			Total Plant Biomass (BIOM)	
PKE ID	Depth (mm)	Hydrological Group	Texture (%)				(kg N/ha Year)			(Metric Tons of Dry Matter/ha Year)	
			Description	Clay	Silt	Sand	Cropland	Grassland	Sum	Cropland	Grassland
1508	1100	B	silty loam	21	66	13	56	6	27	26	9
1536	700	B	silty loam	17	66	17	-	11	11	-	8
1537	950	C	silty loam	24	69	7	57	6	21	25	8
1561	250	D	silty clay loam	37	57	6	118	16	37	17	7
1565	1000	C	loam	14	48	38	68	8	38	25	9
1573	1200	D	silty clay loam	27	64	9	59	7	29	26	9
1577	800	C	silty loam	20	73	7	70	8	36	24	8
1578	920	B	silty loam	15	74	10	71	8	38	26	9
1579	900	C	sandy loam	6	38	55	72	9	40	23	9
1581	1200	B	silty loam	18	62	20	59	6	29	27	9
1583	1200	C	silty loam	16	72	12	54	6	30	27	9
401	1300	B	silty loam	14	79	7	56	6	27	28	9
						Average	64	8	32	25	9

PKE—pedocartographic unit of the soil map of Slovenia (Figure 1).

In contrast, soils with lower clay content (PKE 1579, 1578, 1565) and shallow soils (PKE 1561) exhibit the highest rates of nitrate leaching (ranging from 71 to 118 kg/ha per year) below the soil profile (NO3L). Consequently, stricter fertiliser application restrictions should be enforced on these soil types. Conversely, more lenient measures can be adopted for deeper loamy soils (PKE 1508 and PKE 401).

The crop rotation adaptation scenarios (R2 to R5) have been designed to reduce fertiliser use, introduce winter greening, and expand cover crop management, all of which help to minimise nitrate leaching. Table 6 allows a better understanding of which soils require more targeted adaptations. By implementing measures R1 to R5, we can tailor practices to meet the specific needs of different soil types, ultimately maximising both environmental and economic efficiency.

3.2.2. Nitrate Nitrogen Content in Surface Water at the Outlet of the Subbasin

Table 7 illustrates the variations in total nitrogen (TOT_N) and nitrate nitrogen (NO3_OUT) transported in surface water flow from the subbasins of karst springs. The base scenario (B) represents the average baseline values of total nitrogen from 1998 to 2022. The intensive scenario (R1) indicates an overall increase in nitrogen concentrations across all subbasins, which is attributed to the intensified application of fertilisers. In particular, subbasin 20 exhibits a 35% increase in NO3_OUT.

In contrast, alternative scenarios R1 to R4 demonstrate decreases in total nitrogen, with the most pronounced results occurring in scenario R3. This scenario achieves reductions of up to 51% in TOT_N and up to 32% in NO3_OUT within subbasin 19, which contains the highest proportion of arable land.

Scenarios E1 to E6 present variable effects, with scenario E2 identified as the most effective. This scenario entails a transition from entirely arable land to three-cut grasslands, reducing total nitrate nitrogen by as much as 44% in subbasin 19. Scenarios E4 and E5 exhibit comparable effectiveness.

Table 7. Comparison of results between baseline and alternative scenarios for total nitrogen (TOT_N) and nitrate nitrogen (NO3_OUT) transported by surface water flow from the karst springs subbasins (kg N/year, %).

Simulated Annual Averages (1998–2022)	Subbasin Name and No.										
	Dobličica	**Obršec**	**Jelševnik**	**Janževe Loke**	**Šprajcarjev Zdenec**	**Talački Breg**	**Pački Breg**	**Brežiček**	**Otovski Breg**	**Stobe**	**Planinc**
	1	**8**	**9**	**10**	**11**	**14**	**16**	**17**	**18**	**19**	**20**
Total nitrogen transported out of reach (TOT_N) at subbasin outflow (kg N/year)											
Base model (kg N/year)	34,490	4210	10,511	360	6299	5705	9619	564	7342	267	265
StDv	10,791	1257	3206	106	1812	1837	2794	160	2164	75	82
SCENARIO	Change (%) in the amount of nitrate nitrogen (N-NO_3^-)										
R1	2	4	2	5	13	2	8	11	10	12	14
R2	−2	−6	−4	−18	−14	−2	−17	−22	−17	−34	−23
R3	−2	−8	−5	−20	−19	−3	−20	−26	−21	−39	−28
R4	−4	−11	−7	−26	−28	−4	−27	−36	−29	−51	−39
R5	−3	−10	−6	−23	−24	−4	−24	−31	−25	−45	−34
E1	−1	−2	−3	0	0	−4	−3	0	−3	0	0
E2	−5	−16	−10	−38	−40	−7	−39	−50	−41	−69	−54
E3	1	1	−1	3	2	0	0	2	0	2	5
E4	−3	−13	−7	−34	−35	−2	−34	−45	−36	−65	−46
E5	−6	−18	−14	−37	−37	−9	−37	−47	−39	−66	−51
E6	−2	−8	−5	−21	−18	−3	−15	−24	−16	−36	−6
Nitrate transported with water out of reach (NO3_OUT) at subbasin outflow (kg N/year)											
Base model (kg N/year)	27,911	3041	7321	215	4787	4617	6416	355	4928	124	154
StDv	9620	1081	2715	86	1574	1663	2302	123	1788	45	64
SCENARIO	Change (%) in the amount of nitrate nitrogen (N-NO_3^-)										
R1	2	6	2	9	18	2	14	20	16	35	28
R2	0	−1	−1	−2	−4	−1	−3	−5	−4	−8	−7
R3	−1	−3	−1	−4	−10	−1	−8	−11	−9	−17	−16
R4	−2	−6	−2	−8	−18	−2	−14	−21	−16	−32	−29
R5	−1	−4	−2	−6	−14	−2	−10	−15	−12	−23	−21
E1	0	0	−1	0	0	−1	−1	0	−1	0	0
E2	−3	−8	−3	−11	−26	−3	−19	−29	−22	−44	−40
E3	1	2	1	4	3	2	3	3	3	3	8
E4	−1	−5	−1	−6	−20	1	−14	−22	−16	−36	−27
E5	−2	−7	−3	−9	−22	−2	−16	−25	−18	−38	−33
E6	−1	−3	−1	−2	−7	−1	0	−7	0	−10	19

Scenario descriptions and abbreviations are explained in Table 2; red—increase in N, blue—decrease in N.

In general, the measures implemented in most scenarios demonstrate a high degree of efficacy in subbasins 10–11 and 16–20, which possess a significant proportion of agricultural land. Conversely, in subbasins 1, 8, and 9, where black olm populations are more prevalent and cropland is limited, the impact of the scenarios is minimal.

Scenario E3 assumes that there is only one grass cut per year, influencing the soil's formation and mineralisation of organic matter. This approach can be beneficial as it allows for the accumulation of organic matter, but it also presents specific challenges related to soil management and nutrient utilisation. A notable impact of this scenario is seen in soils currently used for arable land with better quality and fertility. These soils typically have a higher nutrient supply, are deeper, and retain more water, creating ideal conditions for greater biomass production. However, with reduced cutting frequency (only once a year), there may be less nutrient removal from the soil, which could increase the risk of excess nutrients and subsequent nitrogen leaching, particularly in more fertile areas. Over time, grassland species composition is expected to shift, leading to reduced nitrogen leaching. Alternatively, the optimal management of such soils would be to convert them into highly maintained grasslands (for example, those subjected to three cuts per year and additional grass sowing), which could mitigate the risk of excessive nutrient build-up in soil and nitrogen leaching. Adjusting agricultural practices in these regions could significantly enhance the balance between biomass production and the protection of water resources. The impacts of newly introduced measures may be time delayed due to specific soils, hydrology, and biochemical characteristics of the specific karst system [21,41].

Scenario E6 proposes converting permanent grasslands on flat terrain (slope < 7%) into cropland while simultaneously implementing an expanded crop rotation on the remaining cropland (R5). With measures outlined in scenario E6, we do not expect any increase in nitrogen levels in surface water, except for subbasin 19, where most agricultural land is on flat terrain. Scenario E5 demonstrates that converting all arable land to single-cut grassland and transforming all grassland into forests results in significantly lower nitrogen levels only in subbasins with a higher percentage of arable land (specifically, subbasins 16, 18, and 19). Additionally, it is important to highlight that the measures in scenario R4, which involve maintaining the current land use while reducing fertilisation rates by 20%, introducing winter greening, and extending the crop rotation to six years with an additional two years of cover crop management (CGM) cultivation, are similarly effective.

3.3. Evaluating the Potential for Improving Black Olm Habitat by Adapting Agricultural Practices

Various measures are needed when adapting agricultural practices to protect the black olm karst groundwater habitat, which must be implemented in a coordinated and spatially optimised manner to be effective and contribute to improving the habitat quality. It is essential to balance environmental protection with the maintenance of agricultural activity [17]. Particular attention should be given to adapting agricultural production technologies, such as reducing fertiliser use, expanding crop rotations, and implementing cover crops and sustainable grassland management practices. These approaches can significantly reduce nitrogen leaching into groundwater and surface water while maintaining agricultural productivity.

This study indicates that the soil and hydrological systems in karst areas, characterised by rapid infiltration and short water retention times during precipitation, possess unique features that must be considered when managing nitrogen [6,18]. Some measures aimed at protecting drinking water sources in Slovenia have already received financial support under the Strategic Plan of the Common Agricultural Policy (CAP) for 2023–2027 [25]. Through the current CAP agri-environment-climate payments, farmers can opt for interventions that preserve and improve the quality of water sources while enhancing soil properties and fertility. These measures help prevent nitrate leaching by adhering to specific requirements, such as mandatory five-year crop rotations, cultivating at least three different crops, greening arable land with year-round green cover using winter-freeze resistant crops, and restricting the use of phytopharmaceuticals in designated water protection areas.

The CAP also includes regulations related to the Nitrates Directive and water protection area legislation under Conditionality [26]. The "Regulation on the Protection of Waters against Pollution by Nitrates from Agricultural Sources" mandates landowners and land users in Slovenia to implement additional measures for balanced and targeted fertilisation. These include quantitative restrictions on annual nitrogen application, the creation of fertilisation plans, and bans on late autumn and winter fertilisation. These measures effectively reduce nitrate leaching and enhance the quality of water resources. Furthermore, other methods to mitigate nitrate leaching include using cover crops and intercrops in permanent crops, fertilising with composted organic fertilisers, applying smaller doses of fertiliser, utilising mulch or mechanical weed control, and practising conservation tillage.

The evaluation of the impact of various scenarios on the nitrate content in the subbasin outflow demonstrates that a more strategic application of these scenarios could substantially reduce nitrogen leaching and lower nitrate levels in water sources, particularly in areas with a higher share of cropland (Table 8). The benefits of reducing agricultural production intensity, as demonstrated in scenario R4, are also evident in modelling nitrate concentrations at subbasin 11, 17, 19, and 20 outlets, where cropland constitutes a larger portion. By implementing the R4 scenario, we can anticipate a reduction in nitrogen levels to below the threshold (target) value of 9.2 mg NO_3^-/L, which is suitable for the habitat of the black olm.

Table 8. Potential nitrate concentration in the reach of karst spring subbasin outflow upon implementation of the alternative scenarios (R, E).

	Nitrate Concentration in the Reach at Karst Springs Subbasin Outflow (mg NO_3^-/L)										
	Karst Springs Subbasin Name and No.										
	Dobličica	**Obršec**	**Jelševnik**	**Janževe Loke**	**Šprajcarjev Zdenec**	**Talački Breg**	**Pački Breg**	**Brežiček**	**Otovski Breg**	**Stobe**	**Planinc**
	1	**8**	**9**	**10**	**11**	**14**	**16**	**17**	**18**	**19**	**20**
Average observed (ZRC SAZU) (9/2021–11/2022)	3.3	10.1	3.6	4.2	10.3	10.3	13.6	10.2	14.3	12.5	11.1
Target value for the black olm habitat	9.2 mg NO_3^-/L										
Required reduction	-	1.1	-	-	1.3	1.3	4.6	1.2	5.3	3.5	2.1
Scenario	Potential nitrate concentration upon implementation of the scenario										
R1	3.4	10.7	3.7	4.6	12.2	10.5	15.5	12.2	16.6	16.9	14.2
R2	3.3	10.0	3.6	4.1	9.9	10.2	13.2	9.7	13.7	11.5	10.3
R3	3.3	9.8	3.6	4.0	9.3	10.2	12.5	9.1	13.0	10.4	9.3
R4	3.2	9.5	3.5	3.9	8.4	10.1	11.7	8.1	12.0	8.5	7.9
R5	3.3	9.7	3.5	3.9	8.9	10.1	12.2	8.7	12.6	9.6	8.8
E1	3.3	10.1	3.6	4.2	10.3	10.2	13.5	10.2	14.2	12.5	11.1
E2	3.2	9.3	3.5	3.7	7.6	10.0	11.0	7.2	11.2	7.0	6.7
E3	3.3	10.3	3.6	4.4	10.6	10.5	14.0	10.5	14.7	12.9	12.0
E4	3.3	9.6	3.6	3.9	8.2	10.4	11.7	8.0	12.0	8.0	8.1
E5	3.2	9.4	3.5	3.8	8.0	10.1	11.4	7.7	11.7	7.8	7.4
E6	3.3	9.8	3.6	4.1	9.6	10.2	13.6	9.5	14.3	11.3	13.2

white—the limit value of 9.2 mg NO_3^-/L; blue—below the target value, favourable for the habitat; red—above the target value, unfavourable for the habitat.

In subbasins 1, 9, and 10, where there is a significant variation in altitude, and forest land use predominates, the impact of agriculture on water nitrate levels is minimal. The primary nitrogen sources in these areas are the mineralisation of forest residues and atmospheric deposition. In Slovenia, the natural background concentration of nitrogen in water varies by region and specific water body. Generally, the nitrate (NO_3^-) concentration in unpolluted waters is estimated to be around 1 mg N/L (approximately 4.4 mg NO_3^-/L) [24].

Additionally, in subbasins 8, 11, 14, 16, and 18, nitrogen sources may be related to uncontrolled wastewater discharge from residential buildings or inadequately designed storage for organic fertilisers, resulting in unregulated leaching into the groundwater. This conclusion is supported by water quality measurements conducted in the study area within the HAČLORI project [44,45]. Microbial source tracking, utilising molecular and cultivable methods [46] at four selected karst springs (8, 9, 18, and 19), has shown that human activities represent the predominant nitrogen source in the sampled water of subbasin 18. At the same time, subbasins 8 and 9 indicate that human activities are the only additional nitrogen source. Conversely, in subbasin 19, animal sources are identified as the exclusive contributors to nitrogen levels.

To effectively adapt agricultural practices and land use, measures must be implemented in a coordinated manner alongside other initiatives tailored to the specific soil characteristics and karst hydrogeological conditions [16,18,43]. This strategy will facilitate improved nitrogen management and mitigate adverse environmental impacts, which are essential for preserving the black olm habitat and enhancing water quality within its ecosystem.

Karst springs are interconnected with the surface through a complex network of underground channels and tunnels, resulting in a hydraulic continuum that extends from the surface to the springs [16]. This connectivity renders them susceptible to pollution; however, they also play a vital role in sustaining the ecological balance of groundwater ecosystems [6]. Major precipitation events can intensify the geochemical processes inherent in karst geology. Consequently, developing agricultural measures for karst areas necessitates a specialised approach to manage nitrogen balance effectively [17].

The modelling results highlight specific agri-environmental measures that could help reduce nitrogen leaching from agriculture.

(a) These measures include supporting farms in acquiring the knowledge and financial resources needed to transition to sustainable farming practices [17,18,47]. Key recommendations are as follows: (i) Implement a diverse crop rotation that spans at least four years and includes a minimum of three different crops; (ii) Ensure that the soil is covered with cover crops or greening between the main summer or winter crops, preferably using winter resistant cover crops; (iii) Adopt shallow minimum tillage techniques, limiting tillage depth to 10 cm. It is important to note that bare soil without plant cover is more susceptible to nutrient leaching, and ploughing can accelerate the mineralisation of organic matter. Therefore, it is advisable to carry out tillage and sowing within a few days of each other. The proposed measures will involve short-term costs for purchasing seeds for cover crops or additional plants in the rotation. The shift from ploughing to minimum tillage is the most significant expense, which only the largest farmers can typically afford on their own. However, the long-term economic benefits of transitioning to sustainable tillage methods will provide much greater advantages for the environment and improve the productive capacity of agricultural soils.

(b) Including legumes, such as DTM and clover, in the existing crop rotation can help reduce the use of fertilisers. Research from the URAVIVO project has indicated that in some cases, nitrogen fertiliser doses can be reduced by 20% without adversely affecting yields [48,49]. This consideration was incorporated when developing the proposal for the extended R3 scenario rotation. It is recommended that professional services put more effort into promoting crops that require lower nitrogen inputs for growth, such as clover and clover–grass mixes. Raising awareness about the necessity of reducing both the frequency and amount of fertiliser applications while promoting controlled-release fertilisers will yield beneficial economic outcomes for farms. Legumes that require added fertiliser in their production technology, such as fodder peas and soybeans, should not be promoted in the study area to minimise fertiliser use further. Additionally, the mineralisation of underground plant residues can impact N leaching [50].

(c) A comparison of various farming scenarios indicates that cultivation practices involving higher nitrogen (N) inputs can adversely affect the nitrogen balance in the soil, leading to increased nitrogen leaching. It is crucial to note that a single cultivation technology may not be universally effective across different soil types. While a particular practice may have a minimal impact on the nitrogen balance in deeper soils or those with a clay-loam texture, it can result in significantly greater nitrogen leaching in shallower soils. The findings highlight the importance of managing agricultural practices according to specific soil characteristics, such as depth, texture, and skeletalness, to mitigate potential negative nitrogen balances. Developing a precise soil map, ideally at scales of 1:10,000 or 1:5000, grounded in field soil sampling is one of the essential tools to estimate groundwater vulnerability in karst and to implement targeted management [29,42,43].

(d) A comparative analysis of crop rotations following the fertiliser guidelines has revealed that, in certain instances, fertilisers are applied at doses that are technically appropriate but may not correspond to the specific soil types and geological conditions prevalent in the region. It is essential to strengthen oversight in implementing existing measures delineated in the nature protection and groundwater protection legislation and CAP, particularly concerning Conditionality [26,51–53]. To this end, it is recommended that one or more representative experimental plots showcasing diverse soil types be established in critical areas. These plots will enable the evaluation of agri-environmental measures on plant growth, yield, nitrogen leaching, pesticide usage and cost of production. Furthermore, they can function as effective educational tools for farmers, illustrating the practical benefits of these measures on environmental and economic performance. In addition, it is imperative to augment the number of individual consultations available to farmers, focusing on agricultural technologies tailored to each farm's distinctive characteristics.

(e) Organic farming can exhibit similar nitrogen (N) leaching levels as conventional practices when pursuing equivalent yields per hectare. However, in promoting organic farming, it is essential to avoid nitrogen surpluses from fresh organic fertilisers (livestock) and to promote composted manure and green manure. Weather conditions significantly influence nitrogen mineralisation from organic fertilisers and cannot be fully controlled [50,54]. Additionally, soil characteristics, including depth, texture, and water-holding capacity, must be considered when developing fertiliser strategies and agricultural practices.

(f) Intensive grasslands with more frequent cuts show a positive nitrogen (N) balance across various scenarios. Soil properties play a crucial role, especially with manure, slurry, or grazing on shallow soils typical of karst regions [55]. In areas with significant grassland, promoting extensive grazing by herbivores is beneficial. Incentives should support the breeding of suckler cows and small ruminants like sheep and goats, which have lower stocking densities (currently 0.48 livestock units per hectare). This helps improve the distribution of organic fertilisers and nutrient utilisation. Agricultural professionals should focus on educating farmers about grassland management, including aeration, seeding, liming, and proper fertilisation practices. A diverse and dense grassland enhances harvested yield and nutrient use, reduces nutrient losses and minimises environmental impact [56,57]. It is essential to define the type of livestock, stocking density, and appropriate fertilisers with the help of experts, ensuring sustainable environmental and economic outcomes [58].

(g) In the studied area, a significant portion of the land is dedicated to orchards and vineyards, which share similarities with grasslands due to their inter-row cultivation type. From a nitrogen balance perspective, fruit farming is a favourable option. Over recent decades, fertiliser application methods in fruit and grape cultivation have evolved, driven by new insights into the adverse effects of excessive fertilisation on crop quality and storage capacity [59,60]. However, viticulture and fruit growing face challenges related to irrigation and pesticide use. Ensuring a reliable water source is crucial for achieving optimal yield in quantity and quality. However, extracting water from subterranean aquifers could further strain the habitat of the black olm. Phytopharmaceuticals, including fungicides, insecticides, and herbicides, play a vital role in producing fruits and grapes. Therefore, it is essential to manage pesticide application effectively, utilising professional services that guide growers toward sustainable practices. These practices should emphasise preventive measures such as repellents, disruptors, and baits. To evaluate these measures, it is important to assess their impact on subterranean habitats and sensitive species, including the black olm.

Studies on urban areas in the study region [44,45] found that agricultural activities contribute between 50% and 90% of the nitrogen to groundwater within flatter, agriculturally active subbasins. In key karst springs (subbasins 8 and 9), which host the largest populations of the black olm, the contributions from agriculture and unregulated urban wastewater systems are each 50%. To protect the black olm habitat, it is essential to implement measures in both agriculture and residential areas. Specifically, residential buildings should be equipped with waterproof septic tanks, and a public wastewater discharge system should be established to direct effluent to treatment facilities. Additionally, maintaining records of wastewater devices, ensuring regular wastewater removal, and monitoring the integrity of septic tanks are critical for success. Studies on karst water quality from around the world propose that the upstream human populations must be advised of the risks resulting from their daily activities related to wastewater management [18,21,23,39,41–43].

Ensuring the safety and conservation of the endemic black olm (*Proteus anguinus parkelj*) is vital for maintaining the health of the subterranean karst aquatic ecosystem in the studied region. This species acts as an indicator of groundwater quality and overall ecosystem health. Protecting the black olm aids in preserving the unique biodiversity of karst environments, home to many specialised and often endemic species that rely on the same habitat, such as various species of stygobiont amphipods (i.e., *Niphargus* spp.) [10]. Additionally, the black olm supports ecosystem stability by maintaining food web balance. Its presence and health reflect the condition of the ecosystem, and its conservation can have cascading effects on other species. The black olm also provides valuable scientific insights into evolutionary biology, genetics, and ecology due to its unique adaptations to a subterranean lifestyle [10].

Beyond ecological importance, the black olm holds cultural and educational significance, promoting environmental awareness and stewardship. It symbolises the natural heritage of Slovenia and the Dinaric Karst region. By focusing on its conservation, we benefit numerous other species and the broader environment, fostering a sense of stewardship for natural resources [61].

When evaluating the impact of the scenarios, several uncertainties associated with the establishment of the model were duly considered. These uncertainties arise from various factors, including production technologies, crop rotations, harvest schedules, mowing dates, machine operation timelines, and fertiliser application practices. As a result, the outcomes produced by the model represent average estimates; individual farmers adapt their production schedules and technologies to accommodate specific crops, animal species, production intensities, and fluctuating weather conditions.

The final recommendations, formulated based on the model scenarios, should be interpreted as potential ecosystem responses to alterations in land use and agricultural technologies, as well as the anticipated effects of agri-environmental measures on the nitrogen balance within soil and water resources. The results and recommendations should be used for the management of karst areas in an interdisciplinary way, involving science as well as socio-economic and socio-political domains [62]

4. Conclusions

This research emphasises the significant impact of agricultural practices on soil and groundwater resources in karst areas, particularly within the unique habitat of the black olm. The water dynamics in this region are complex due to karst geology, making modelling challenging. Our scenarios demonstrated that adjusting crop rotation (R2, R4), fertilisation (R3), and land use (E2, E4, E5) can effectively reduce nitrate leaching into groundwater while considering environmental and socio-economic factors. This is vital for maintaining water quality, safeguarding endemic species like the black olm, and ensuring future sustainable coexistence with human populations. Furthermore, the results indicate that the target value of 9.2 mg NO_3^-/L cannot be achieved solely through agricultural measures; urban point sources must also be managed.

However, it is important to acknowledge some limitations and uncertainties in our research. One key area for improvement is the limited availability of accurate data on soil physical properties, which could impact the precision of our simulations. Additionally, the modelling is based on certain assumptions that may not fully capture the variability of natural processes in karst systems. To enhance prediction accuracy, it would be beneficial to incorporate a larger number of measurements, such as flow discharge and nitrogen content in the water of Karst spring, to extend the model calibration and validation period.

This research offers the scientific community a new approach to modelling subterranean karst aquifers and understanding how agricultural practices affect them. This knowledge is crucial for future research and managing environmentally sensitive subterranean groundwater habitats. For professionals, particularly decision-makers and agricultural policy planners, our findings provide concrete guidelines for directing agricultural practices that can mitigate adverse impacts on groundwater and surface water quality.

In the future, we could focus on refining models to predict the effects of climate change on karst aquifers, particularly concerning extreme weather events like droughts and floods, and evaluate their influence on groundwater resources and biodiversity.

Future efforts should also aim to predict the effects of climate change, analyse long-term agri-environmental measures, and assess the impact of urbanisation on karst ecosystems and water resources. An important aspect to explore is the socio-economic effects of proposed measures as part of environmental policies on local rural communities, where agriculture and land use are closely linked to natural resource conservation and settlement.

Finally, interdisciplinary studies examining the relationship between human activities, biodiversity, and groundwater quality could enhance our understanding of how biodiversity in karst regions contributes to natural water purification, thereby improving groundwater quality. Such research could inform more sustainable management of karst ecosystems and their water resources.

Author Contributions: Conceptualisation, M.G. and R.C.; methodology, M.G.; software, M.G.; validation, M.G.; formal analysis, M.G.; investigation, M.G. and R.C.; resources, M.G.; data curation, M.G.; writing—original draft preparation, M.G.; writing—review and editing, R.C.; visualisation, M.G.; supervision, R.C.; project administration, R.C.; funding acquisition, R.C. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by the Slovenian Research and Innovation Agency, Ministry of Natural Resources and Spatial Planning and Ministry of Agriculture, Forestry and Food, grant number CRP 2021 V1-2139 (HAČLORI).

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: The raw data supporting the conclusions of this article will be made available by the authors on request.

Conflicts of Interest: The authors declare no conflicts of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results.

References

1. Culver, D.C.; Pipan, T. *The Biology of Caves and Other Subterranean Habitats*, 2nd ed.; Biology of habitats series; Oxford Academic: Oxford, UK, 2019. [CrossRef]
2. Zagmajster, M.; Malard, F.; Eme, D.; Culver, D.C. Subterranean Biodiversity Patterns from Global to Regional Scales. In *Cave Ecology*; Moldovan, O.T., Kováč, L., Halse, S., Eds.; Springer: Berlin/Heidelberg, Germany, 2018; pp. 195–227. [CrossRef]
3. Bregović, P.; Zagmajster, M. Understanding hotspots within a global hotspot—Identifying the drivers of regional species richness patterns in terrestrial subterranean habitats. *Insect Conserv. Divers.* **2016**, *9*, 268–281. [CrossRef]
4. Aljančič, G. History of research on *Proteus anguinus* Laurenti 1768 in Slovenia/Zgodovina raziskovanja človeške ribice (Proteus anguinus Laurenti 1768) v Sloveniji. *Folia Biol. Geol.* **2019**, *60*, 39–69. [CrossRef]
5. The Black Olm/Črna Človeška Ribica. Available online: https://repozitorij.uni-lj.si/IzpisGradiva.php?id=37125&lang=eng (accessed on 8 September 2024).
6. Koren, K.; Brajkovič, R.; Bajuk, M.; Vraničar, Š.; Fabjan, V. Hydrogeological characterisation of karst springs of the white (*Proteus anguinus* anguinus) and black olm (*Proteus anguinus* parkelj) habitat in Bela krajina (SE Slovenia). *Geologija* **2023**, *66*, 151–166. [CrossRef]
7. Mezga, K.; Janža, M.; Prestor, J.; Koren, K.; Šram, D. Groundwater dependent ecosystems—Groundwater status indicators. *Nat. Slov.* **2016**, *18*, 35–42. [CrossRef]
8. Jensen, K.M.; Jensen, M.H.; Kristensen, E. Nitrification and denitrification in Wadden Sea sediments (Königshafen, Island of Sylt, Germany) as measured by nitrogen isotope pairing and isotope dilution. *Aquat. Microb. Ecol.* **1996**, *11*, 181–191. [CrossRef]
9. Scott, G.; Crunkilton, R.L. Acute and chronic toxicity of nitrate to fathead minnows (*Pimephales promelas*), ceriodaphnia dubia, and Daphnia magna. *Environ. Toxicol. Chem.* **2000**, *19*, 2918–2922. [CrossRef]
10. Mali, L.B.; Sket, B. History and biology of the «black proteus« (*Proteus anguinus parkelj* Sket & Arntzen 1994; Amphibia: Proteidae): A review/Zgodovina in biologija črnega močerila (*Proteus anguinus parkelj* Sket & Arntzen 1994; Amphibia: Proteidae): Pregledni članek. *Folia Biol. Geol.* **2019**, *60*, 5–37. [CrossRef]

11. Kolar, B. The threshold concentration for nitrate in groundwater as a habitat of Proteus anguinus. *Nat. Slov.* **2018**, *20*, 39–42. [CrossRef]
12. Palmer, A.N. Geomorphic interpretation of karst features. In *Groundwater as a Geomorphic Agent*; Routledge: London, UK, 1984; pp. 173–209. [CrossRef]
13. Lisec, A.; Montanarella, L.; Vrščaj, B.; Kralj, T.; Lobnik, F.; Suhadolc, M.; Prus, T.; Mihelič, R.; Rupreht, J.; Zupan, M.; et al. Tla Slovenije s Pedološko Karto v Merilu 1:250000 = Soils of Slovenia with Soil Map 1:250000. European Commission Joint Reaearch Centre (JRC): Publications Office of the European Union, Luxembourg. 2005. Available online: https://op.europa.eu/sl/publication-detail/-/publication/538dee5b-dfbf-45d6-bc3c-d4b6fab3110d (accessed on 25 October 2024).
14. Urbančič, M.; Simončič, P.; Prus, T.; Kutnar, L. Atlas of Slovenian Forest Soils/Atlas Gozdnih tal Slovenije. Gozdarski Inštitut Slovenije, Ljubljana. 2005. Available online: https://plus.cobiss.net/cobiss/si/sl/bib/223504896 (accessed on 25 October 2024).
15. Tuyet, D. Characteristics of Karst Ecosystems of Vietnam and Their Vulnerability to Human Impact. *Acta Geol. Sin.—Engl. Ed.* **2001**, *75*, 325–329. [CrossRef]
16. Bakalowicz, M. Karst groundwater: A challenge for new resources. *Hydrogeol. J.* **2005**, *13*, 148–160. [CrossRef]
17. Coxon, C. Agriculture and Karst. In *Karst Management*; Beynen, P.E., Ed.; Springer: Berlin/Heidelberg, Germany, 2011; pp. 103–138. [CrossRef]
18. Guo, F.; Yuan, D.; Qin, Z. Groundwater Contamination in Karst Areas of Southwestern China and Recommended Countermeasures. *Acta Carsologica* **2010**, *39*, 389–399. [CrossRef]
19. Zeiger, S.J.; Owen, M.R.; Pavlowsky, R.T. Simulating non-point source pollutant loading in a karst basin: A SWAT modeling application. *Sci. Total Environ.* **2021**, *785*, 147295. [CrossRef]
20. Lukač Reberski, J.; Terzić, J.; Maurice, L.D.; Lapworth, D.J. Emerging organic contaminants in karst groundwater: A global level assessment. *J. Hydrol.* **2022**, *604*, 127242. [CrossRef]
21. Fenton, O.; Mellander, P.E.; Daly, K.; Wall, D.P.; Jahangir, M.M.R.; Jordan, P.; Hennessey, D.; Huebsch, M.; Blum, P.; Vero, S.; et al. Integrated assessment of agricultural nutrient pressures and legacies in karst landscapes. *Agric. Ecosyst. Environ.* **2017**, *239*, 246–256. [CrossRef]
22. Stevanović, Z.; Stevanović, A.M. Monitoring as the Key Factor for Sustainable Use and Protection of Groundwater in Karst Environments—An Overview. *Sustainability* **2021**, *13*, 5468. [CrossRef]
23. Brad, T.; Bizic, M.; Ionescu, D.; Chiriac, C.M.; Kenesz, M.; Roba, C.; Ionescu, A.; Fekete, A.; Mirea, I.C.; Moldovan, O.T. Potential for Natural Attenuation of Domestic and Agricultural Pollution in Karst Groundwater Environments. *Water* **2022**, *14*, 1597. [CrossRef]
24. Quality of Groundwater/Kakovost Podzemne Vode. Available online: https://kazalci.arso.gov.si/sl/content/kakovost-podzemne-vode-1?tid=16 (accessed on 20 October 2024).
25. Nitrates Directive (91/676/EEC). Available online: https://environment.ec.europa.eu/topics/water/nitrates_en (accessed on 20 October 2024).
26. Slovenian Strategic Plan for the Common Agricultural Policy 2023–2027. Available online: https://skp.si/skupna-kmetijska-politika-2023-2027 (accessed on 10 September 2024).
27. Glavan, M.; Cvejić, R.; Zupanc, V.; Vodnik, K.; Pintar, M. *Modeling the Environmental Impact of Agricultural Measures to Protect the Habitat of the Black Mantis (Proteus anguinus) from Nitrate Pollution/Modeliranje Okoljskega Vpliva Kmetijskih Ukrepov za Varovanje Habitata Črne Človeške (Proteus anguinus Parkelj) pred Onesnaženjem z Nitrati*; Biotechnical Faculty, University of Ljubljana: Ljubljana, Slovenia, 2024; Available online: https://plus.cobiss.net/cobiss/si/sl/bib/210724355 (accessed on 20 October 2024).
28. Arnold, J.G.; Moriasi, D.N.; Gassman, P.W.; Abbaspour, K.C.; White, M.J.; Srinivasan, R.; Santhi, C.; Harmel, R.D.; Griensven, A.; van Liew, M.W.; et al. SWAT: Model Use, Calibration, and Validation. *Trans. ASABE* **2012**, *55*, 1491–1508. [CrossRef]
29. Busico, G.; Colombani, N.; Fronzi, D.; Pellegrini, M.; Tazioli, A.; Mastrocicco, M. Evaluating SWAT model performance, considering different soils data input, to quantify actual and future runoff susceptibility in a highly urbanised basin. *J. Environ. Manag.* **2020**, *266*, 110625. [CrossRef]
30. Gassman, P.W.; Sadeghi, A.M.; Srinivasan, R. Applications of the SWAT Model Special Section: Overview and Insights. *J. Environ. Qual.* **2014**, *43*, 1–8. [CrossRef] [PubMed]
31. Čerkasova, N.; Mėžinė, J.; Idzelytė-, R.; Lesutienė, J.; Ertürk, A.; Umgiesser, G. Exploring variability in climate change projections on the Nemunas River and Curonian Lagoon: Coupled SWAT and SHYFEM modeling approach. *Ocean. Sci.* **2024**, *20*, 1123–1147. [CrossRef]
32. Plunge, S.; Schürz, C.; Čerkasova, N.; Strauch, M.; Piniewski, M. SWAT+ model setup verification tool: SWATdoctR. *Environ. Model. Softw.* **2024**, *171*, 105878. [CrossRef]
33. Zhang, Z.; Chen, S.; Wan, L.; Cao, J.; Zhang, Q.; Yang, C. The effects of landscape pattern evolution on runoff and sediment based on SWAT model. *Environ. Earth Sci.* **2021**, *80*, 2. [CrossRef]
34. Saxton, K.E.; Rawls, W.J. Soil Water Characteristic Estimates by Texture and Organic Matter for Hydrologic Solutions. *Soil Sci. Soc. Am. J.* **2006**, *70*, 1569–1578. [CrossRef]
35. Saxton, K.E.; Rawls, W.J.; Romberger, J.S.; Papendick, R.I. Estimating Generalized Soil-water Characteristics from Texture. *Soil Sci. Soc. Am. J.* **1986**, *50*, 1031–1036. [CrossRef]
36. Abbaspour, K.C.; Vaghefi, S.A.; Srinivasan, R. A Guideline for Successful Calibration and Uncertainty Analysis for Soil and Water Assessment: A Review of Papers from the 2016 International SWAT Conference. *Water* **2018**, *10*, 6. [CrossRef]

37. Moriasi, D.N.; Arnold, J.G.; Liew MW van Bingner, R.L.; Harmel, R.D.; Veith, T.L. Model Evaluation Guidelines for Systematic Quantification of Accuracy in Watershed Simulations. *Trans. ASABE* **2007**, *50*, 885–900. [CrossRef]
38. Moriasi, D.N.; Gitau, M.W.; Pai, N.; Daggupati, P. Hydrologic and Water Quality Models: Performance Measures and Evaluation Criteria. *Trans. ASABE* **2015**, *58*, 1763–1785. [CrossRef]
39. Binet, S.; Joigneaux, E.; Pauwels, H.; Albéric, P.; Fléhoc, C.; Bruand, A. Water exchange, mixing and transient storage between a saturated karstic conduit and the surrounding aquifer: Groundwater flow modeling and inputs from stable water isotopes. *J. Hydrol.* **2017**, *544*, 278–289. [CrossRef]
40. Rusjan, S.; Sapač, K.; Petrič, M.; Lojen, S.; Bezak, N. Identifying the hydrological behavior of a complex karst system using stable isotopes. *J. Hydrol.* **2019**, *577*, 123956. [CrossRef]
41. Wang, Z.J.; Yue, F.J.; Lu, J.; Wang, Y.C.; Qin, C.Q.; Ding, H.; Xue, L.L.; Li, S.L. New insight into the response and transport of nitrate in karst groundwater to rainfall events. *Sci. Total Environ.* **2022**, *818*, 151727. [CrossRef]
42. Moreno-Gómez, M.; Liedl, R.; Stefan, C.; Pacheco, J. Theoretical analysis and considerations of the main parameters used to evaluate intrinsic karst groundwater vulnerability to surface pollution. *Sci. Total Environ.* **2024**, *907*, 167947. [CrossRef] [PubMed]
43. Bakalowicz, M. Management of Karst Groundwater Resources. In *Karst Management*; Beynen, P.E., Ed.; Springer: Berlin/Heidelberg, Germany, 2011; pp. 263–282. [CrossRef]
44. Research to Identify and Prevent Pollution of the Catchment Hinterland of Jelševniščica and Otovski breg, with Special Attention to the Habitat of the Black Olm/Raziskave za opredelitev in preprečevanje obremenjevanja vodozbirnega zaledja Jelševniščice in Otovskega brega, s posebnim ozirom na habitat črne človeške ribice. ZRC SAZU, Ljubljana. 2024. Available online: https://izrkp.zrc-sazu.si/sl/programi-in-projekti/raziskave-za-opredelitev-in-preprecevanje-obremenjevanja-vodozbirnega-zaledja (accessed on 5 November 2024).
45. Prelovšek, M.; Curk, M.; Mali, N.; Mulec, J.; Petrič, M.; Pintar, M.; Urbanc, J.; Cvejić, R. Research to identify and prevent pollution of the catchment hinterland of Jelševniščica and Otovski breg, with special attention to the habitat of the black olm/Raziskave za opredelitev in preprečevanje obremenjevanja vodozbirnega zaledja Jelševniščice in Otovskega brega, s posebnim ozirom na habitat črne človeške ribice. In Proceedings of the 33. Mišičev Vodarski Dan 2022, Maribor, Slovenia, 5 October 2022; Available online: https://www.mvd20.com/LETO2022/R16.pdf (accessed on 20 October 2024).
46. Sresung, M.; Paisantham, P.; Ruksakul, P.; Kongprajug, A.; Chyerochana, N.; Gallage, T.P.; Srathongneam, T.; Rattanakul, S.; Maneein, S.; Surasen, C.; et al. Microbial source tracking using molecular and cultivable methods in a tropical mixed-use drinking water source to support water safety plans. *Sci. Total Environ.* **2023**, *876*, 162689. [CrossRef] [PubMed]
47. Guo, F.; Jiang, G. Karst Groundwater Management through Science and Education. *Open J. Geol.* **2011**, *1*, 45–50. [CrossRef]
48. Pintar, M.; Arh Marinčič, Š.; Cerar, S.; Cesar, A.; Curk, M.; Cvejić, R.; Glavan, M.; Kastelec, D.; Koroša, A.; Korpar, P.; et al. *Efficient Use of Water and Nutrients in Crop Production to Protect and Improve Drinking Water Resources: URaViVO: Final Report/Učinkovitejša Raba Vode in Hranil v Rastlinski Pridelavi za Varovanje in Izboljšanje Virov Pitne Vode: URaViVO: Končno Poročilo*; Biotechnical Faculty, University of Ljubljana: Ljubljana, Slovenia, 2021; Available online: https://plus.cobiss.net/cobiss/si/sl/bib/101641475 (accessed on 1 November 2024).
49. Curk, M.; Glavan, M. Assessing and Mapping the Environmental Impacts of Best Management Practices in Nitrate-Vulnerable Areas. *Water* **2023**, *15*, 2364. [CrossRef]
50. Benoit, M.; Garnier, J.; Billen, G.; Tournebize, J.; Gréhan, E.; Mary, B. Nitrous oxide emissions and nitrate leaching in an organic and a conventional cropping system (Seine basin, France). *Agric. Ecosyst. Environ.* **2015**, *213*, 131–141. [CrossRef]
51. Giordano, R.; D'Agostino, D.; Apollonio, C.; Scardigno, A.; Pagano, A.; Portoghese, I.; Lamaddalena, N.; Piccinni, A.F.; Vurro, M. Evaluating acceptability of groundwater protection measures under different agricultural policies. *Agric. Water Manag.* **2015**, *147*, 54–66. [CrossRef]
52. Goldscheider, N. A holistic approach to groundwater protection and ecosystem services in karst terrains. *Carbonates Evaporites* **2019**, *34*, 1241–1249. [CrossRef]
53. Ravbar, N.; Šebela, S. The effectiveness of protection policies and legislative framework with special regard to karst landscapes: Insights from Slovenia. *Environ. Sci. Policy* **2015**, *51*, 106–116. [CrossRef]
54. Pandey, A.; Li, F.; Askegaard, M.; Rasmussen, I.A.; Olesen, J.E. Nitrogen balances in organic and conventional arable crop rotations and their relations to nitrogen yield and nitrate leaching losses. *Agric. Ecosyst. Environ.* **2018**, *265*, 350–362. [CrossRef]
55. Valkama, E.; Rankinen, K.; Virkajärvi, P.; Salo, T.; Kapuinen, P.; Turtola, E. Nitrogen fertilisation of grass leys: Yield production and risk of N leaching. *Agric. Ecosyst. Environ.* **2016**, *230*, 341–352. [CrossRef]
56. Egan, G.; McKenzie, P.; Crawley, M.; Fornara, D.A. Effects of grassland management on plant nitrogen use efficiency (NUE): Evidence from a long-term experiment. *Basic Appl. Ecol.* **2019**, *41*, 33–43. [CrossRef]
57. Leimer, S.; Oelmann, Y.; Wirth, C.; Wilcke, W. Time matters for plant diversity effects on nitrate leaching from temperate grassland. *Agric. Ecosyst. Environ.* **2015**, *211*, 155–163. [CrossRef]
58. Klaus, V.H.; Friedritz, L.; Hamer, U.; Kleinebecker, T. Drought boosts risk of nitrate leaching from grassland fertilisation. *Sci. Total Environ.* **2020**, *726*, 137877. [CrossRef] [PubMed]
59. Brunetto, G.; Oliveira, B.S.; Ambrosini, V.G.; Couto, R.d.R.; Sete, P.B.; dos Santos Junior, E.; Loss, A.; da Silva, L.O.S.; Gatiboni, L.C. Nitrogen availability in an apple orchard with weed management. *Ciência Rural* **2018**, *48*, e20160895. [CrossRef]
60. He, Z.; Hu, Q.; Zhang, Y.; Cao, H.; Nan, X. Effects of irrigation and nitrogen management strategies on soil nitrogen and apple yields in loess plateau of China. *Agric. Water Manag.* **2023**, *280*, 108220. [CrossRef]

61. Sket, B. Discovering the black proteus *Proteus anguinus* parkelj (Amphibia: Caudata). *Nat. Slov.* **2017**, *19*, 27–28. [CrossRef]
62. Kosič Ficco, K.; Sasowsky, I.D. An interdisciplinary framework for the protection of karst aquifers. *Environ. Sci. Policy* **2018**, *89*, 41–48. [CrossRef]