

# Effect of invasive black locust (*Robinia pseudoacacia* L.) on nitrogen cycle in floodplain ecosystem



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## ABSTRACT

Black locust colonization of floodplains can exacerbate the problems associated with increasing nitrogen inputs into the riparian area and river ecosystem. In this study we compare the nitrogen budgets of two sites of floodplain ecosystem, one is colonized by invasive black locust and the other is forested by the indigenous willow species. Our data report the considerably higher N-flow rates, nitrogen storage and the faster total N-related ecosystem processes in the *Robinia* site in comparison to the indigenous *Salix* site of the study floodplain. Black locust affects the nitrogen cycle in the study riparian ecosystem through the N-fixation, high N-content litterfall and rapid litter decomposition that results in the increase of the nitrogen flow into the river and contaminates the water. Black locust appeared to reduce the conservation values of colonized areas and adjacent river. Restoration strategies that remove the black locust from invaded sites and prevent the further invasion will slow down the contamination. However, high availability of nitrogen in soil and soil degradation might have a long-term effect on floodplain ecosystem after the restoration.

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## 1. Introduction

Black locust (*Robinia pseudoacacia* L.) is one of the most abundant tree in the world after the Eucalyptus and Poplar (Boring and Swank, 1984). It is cultivated worldwide because of the high-quality wood properties, high tolerance against drought and erosion (Böhm et al., 2011). In Ukraine black locust is planted as a roadside tree, an erosion-control species, as well as the tree species for the vegetation buffer zone around the industrial facilities. It is highly competitive to native species as it grows efficiently on poor-nutrient soils and adapts easily to early successional conditions and rapidly invades indigenous communities (Rice et al., 2004). *Robinia* is planted to prevent erosion on the highland floodplains of the rivers in Ukraine, where it moves fast to the riparian area.

Black locust colonization of riparian ecosystems can exacerbate the problems associated with increasing nitrogen inputs into the floodplain area and river ecosystem. Many studies have reported a high capacity of *Robinia pseudoacacia* for the dinitrogen fixation

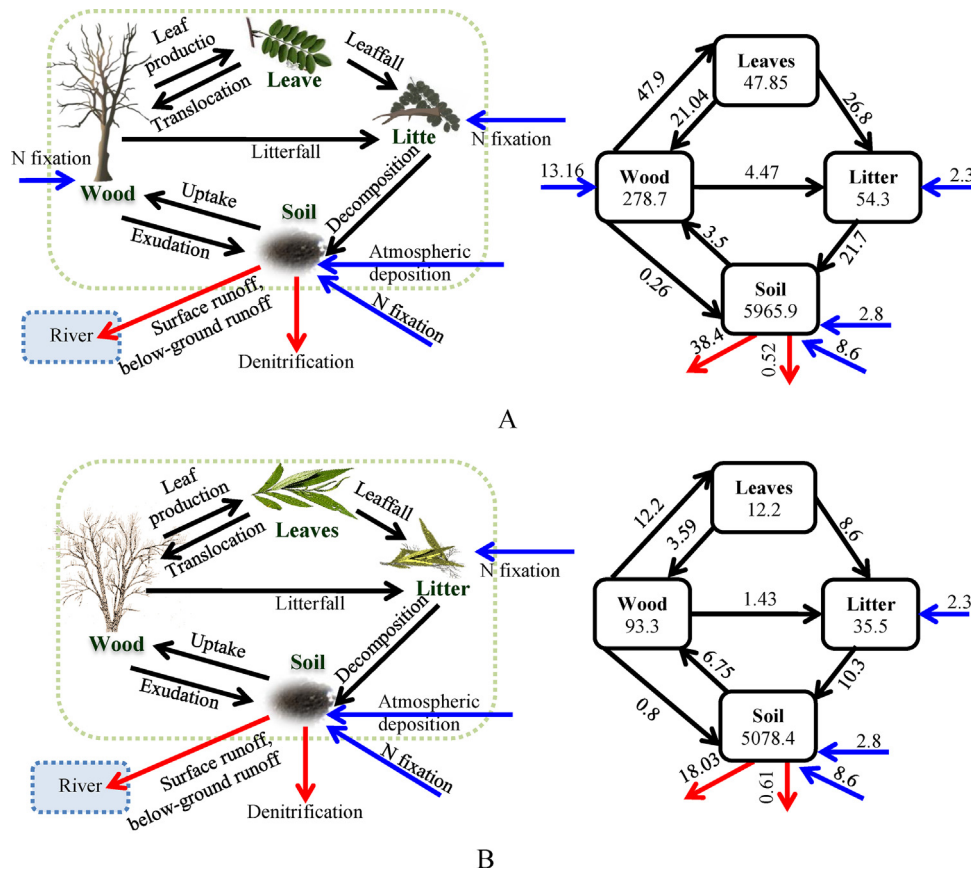
(Boring and Swank, 1984; Danso et al., 1995). High amount of fixed nitrogen may exceed the demand for plant nutrition causing the excessed nitrogen to accumulate in soil and to lose through leaching, runoff and denitrification (Berthold, 2005). These processes may be followed by soil degradation and loss of base cations causing a significant decrease in the pH and increase in the soil exchangeable Al (Van Miegroet and Cole, 1984; Berthold, 2005). Furthermore, frequent inundation of floodplain can cause the nitrate contamination of river water. Leaching of nitrate can lead also to the nitrate pollution of ground water (Berthold, 2005).

Several studies have reported that rates of litter decomposition and N mineralization under *Robinia* stands are higher than under nitrogen non-fixing trees (Tateno et al., 2007; Malcolm et al., 2008) causing strong N-enrichment of soil and subsequent invasion by non-native species and loss of biodiversity (Levine et al., 2003).

More than a decade the black locust has colonized the riparian areas of Prut River in Ukraine. Such invaded sites of the floodplain represent a potential source of nitrogen flow into the Prut River.

The aim of the current study was to investigate the effect of the black locust on nitrogen budget in the riparian area (lowland floodplain) of the Prut River in Chernivtsi Region in Ukraine. To understand the impact of the *Robinia* on the study ecosystem we

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**Fig. 1.** Conceptual diagrams (left) and the parametrized models (right) of the nitrogen budgets of the black locust (A) and willow (B) study sites of Prut river floodplain in Ukraine.

compared two sites of the study floodplain, one colonized by the invasive black locust and the other forested by the indigenous willow species.

The specific objectives of this study were as follows:

- (i) To measure and analyze the nitrogen related ecosystem processes within the each of the study site.
- (ii) To build the nitrogen budget models for each of the study systems and to compare the N-flow rates, N-storage, cycling and system N turnover time among the *Robinia* colonized site and the *Salix* spp. planted area.
- (iii) To investigate the soil properties in each of the study sites in order to elucidate the *Robinia*-mediated changes in the soil.

The conceptual model diagrams for each of the study systems are presented in Fig. 1.

## 2. Materials and methods

### 2.1. Study area

The data for this study were collected from two study sites of the floodplain of river Prut within the village boundaries of Boyany, in Chernivtsi Region in Ukraine (with average sea level 158 m) (Fig. 2). Prut River (approximately 850 km long) originates on the eastern slope of Mountain Hoverla in the Carpathian Mountains and flows through Romania and Moldova, joining the Danube River. The study floodplain is inundated annually for 2–3 months during late March – April.

The study site I (48°16'59.5" N 26°08'12.7" E) is near 50 m wide and it is colonized along the stream with the black locust

stands (*Robinia pseudoacacia* L.) near 12 years old. The study site II (48°16'18.7" N 26°07'21.2" E) is forested with the different age indigenous willow species (*Salix* spp.), namely *Salix caprea* L. and *Salix alba* L.

### 2.2. Sampling and analysis

At each of the two sites the sampling was conducted along three 100 m<sup>2</sup> plots (transects), placed parallel to one another along the river. Most of the field work was conducted during the lowest river water level within the summer period during June–July, 2013. The peak of the plant vegetation period was June 21.

#### 2.2.1. Plant biomass and N content

Each tree within the study area was examined for the shoot basal diameter (SBD) using the forestry caliper about 10 cm and 15 cm above soil for the *Robinia* and *Salix* spp. respectively. Wood (trunk and branches) and leaves dry weight for each *Robinia* tree were calculated using the biomass-SBD allometric equations by Boring and Swank (1984), as follows:

$$y_{\text{leaves}} = 0.0293764965196153 \times x^{1.708}$$

$$y_{\text{branches}} = 0.0246036760414763 \times x^{2.321}$$

$$y_{\text{trunk}} = 0.0246036760414763 \times x^{2.901}$$

where *y* is an oven-dried weight (kg), *x* is diameter (cm) at 10 cm above-ground level.



**Fig. 2.** Location of the study area (river Prut floodplain, Boyany, Chernivtsi Region, Ukraine). Study site I is colonized by *Robinia pseudoacacia* L.; study site II is forested with *Salix* spp.

For *Salix* spp. tree biomass was calculated using allometric biomass equations by [Smith and Brand \(1983\)](#):

$$y_{\text{leaves}} = 12.28 \times x^{2.12}$$

$$y_{\text{wood}} = 43.316 \times x^{2.726}$$

where  $y$  is an oven-dried weight (g),  $x$  is diameter (cm) at 15 cm above-ground level.

Leaves and wood mass standing stocks ( $\text{kg ha}^{-1}$ ) were estimated based on the tree density within each plot.

Fresh leaves and wood (branches and bark parts together) were sampled from four trees within each of the study plot and pooled within the plot into the three replicates separately for leaves and for wood fractions. Samples were oven dried at  $60^\circ\text{C}$  for 48 h, weighted and analyzed for the total N using the Kjeldahl method and colorimetric detection (photocolorimeter КФК-3; EuroLab, St Petersburg, Russia). Leaves and wood nitrogen standing stocks ( $\text{kg ha}^{-1}$ ) were estimated by multiplying leaves and wood mass ( $\text{kg ha}^{-1}$ ) respectively by their N content.

### 2.2.2. Litter sampling and analysis

Litter was sampled from the forest floor using the wood frame of ( $25 \times 25$ )  $\text{cm}^2$  inner size ( $625 \text{ cm}^2$ ). Litter sampling was conducted in four replicates of randomly selected plots within each transect. Samples were oven dried at  $60^\circ\text{C}$  for 48 h and weighted.

### 2.2.3. Litterfall traps experiment

Litterfall was collected in 12-month period of 2013 using three  $1 \text{ m}^2$  litterfall traps for each plot. Litterfall was sorted on fractions of fallen leaves and woody parts (branches and bark parts), oven-dried at  $60^\circ\text{C}$  for 48 h, melted into powder and analyzed for total N using the Kjeldahl digestion method and colorimetric detection (photocolorimeter КФК-3; EuroLab, St Petersburg, Russia). We assume that annual leaf fall biomass ( $\text{kg ha}^{-1} \text{ y}^{-1}$ ) is equivalent to the leaf production biomass ( $\text{kg ha}^{-1} \text{ y}^{-1}$ ) and equal to the allometrically measured leaves biomass ( $\text{kg ha}^{-1}$ ). Nitrogen flow due to the leaf fall was estimated by multiplying the annual leaf fall biomass ( $\text{kg ha}^{-1} \text{ y}^{-1}$ ) by nitrogen content in fallen leaves ( $\text{kg kg}^{-1}$ ). Nitrogen flow due to the translocation (i.e. movement of nutrients back to the wood before the leaf fall) was estimated as the difference in the annual N leaf production and annual N leaf fall. Fallen wood biomass ( $\text{kg ha}^{-1} \text{ y}^{-1}$ ) was measured based on the

litterfall trap experiment on an annual basis. Nitrogen flow due to the fallen wood was estimated by multiplying the fallen wood biomass ( $\text{kg ha}^{-1} \text{ y}^{-1}$ ) by total nitrogen content in wood ( $\text{kg kg}^{-1}$ ).

### 2.2.4. Litter decomposition experiment

Litter decomposition rates were measured using litterbag experiment. Litterbags ( $10 \times 10 \text{ cm}$ ) filled with the site-specific oven-dried litter (5 g) were placed in two replicates within each plot for 30 days in March, June and November in 2013. Litterbags were oven-dried and weighted. Litter decomposition ( $\text{kg ha}^{-1} \text{ y}^{-1}$ ) was estimated using the decomposition rates and litter biomass ( $\text{kg ha}^{-1}$ ). Nitrogen flow due to the litter decomposition was estimated by multiplying the litter decomposed biomass by nitrogen content in litter.

### 2.2.5. Soil sampling and analysis

Soil cores ( $25 \times 25$ )  $\text{cm}^2$  in 20 cm depth (core volume  $12,500 \text{ cm}^3$ ) were sampled from four randomly selected plots within each transect at each of the two sites. Samples from all plots were pulled, mixed and oven dried at  $60^\circ\text{C}$  for 48 h. Samples were sieved and weighted. Soil bulk density was computed as a ratio of the soil oven-dried weight to core volume. Soil mass ( $\text{kg ha}^{-1}$ ) was estimated using soil bulk density.

Soil organic carbon was measured using Tjurin method modified by Kononova, 1966, as follows: soil samples were dichromate digested with 10 ml 0.4 N oxidizing solution ( $\text{K}_2\text{Cr}_2\text{O}_4:\text{H}_2\text{SO}_4$ -1:1) by applying external heat of  $150^\circ\text{C}$  for 20 min in presence of catalyst  $\text{Ag}_2\text{SO}_4$ ; the consumed oxidizing agent was determined by titration with  $\text{Fe}(\text{NH}_4)_2(\text{SO}_4) \cdot 6\text{H}_2\text{O}$  using phenylanthranilic acid as an indicator.

The soil pH was determined by standard glass electrode pH-meter using a potassium chloride solution in 1:2 ratio (soil: 0.1 N KCl).

Soil total N content was measured using the Kjeldahl digestion method and colorimetric detection (photocolorimeter КФК-3; EuroLab, St Petersburg, Russia). The soil N standing stock ( $\text{kg ha}^{-1}$ ) was estimated by multiplying the soil mass ( $\text{kg ha}^{-1}$ ) by the total soil N content ( $\text{kg kg}^{-1}$ ).

### 2.2.6. Atmospheric deposition

Annual precipitation was measured using the pluviograph of  $500 \text{ cm}^2$ . The average annual precipitation rate was 600 mm per  $500 \text{ cm}^2$  that is  $6,000,000 \text{ l h}^{-1} \text{ y}^{-1}$ . Nitrogen input by precipitation

( $\text{kg h}^{-1} \text{y}^{-1}$ ) was estimated by multiplying the annual precipitation by N content in precipitation water ( $\text{kg l}^{-1}$ ).

#### 2.2.7. Above-ground and below-ground water run-off

The long-term measurements of Solovey (2004) were used to calculate the above- and below-ground water inflow into the river Prut. Solovey (2004) shows that the average module of total water inflow into the river Prut is  $10.81 \text{ s}^{-1} \text{ km}^{-2}$ . In accordance with that the annual total water flow ( $V_w$  [ $\text{lh}^{-1} \text{y}^{-1}$ ]) was calculated ( $V_w = 3,405,888 \text{ lh}^{-1} \text{y}^{-1}$ ). Annual nitrogen outflow from soil compartment ( $i$ ) to the river Prut ( $y_i$ ) was estimated as follows:

$$y_i = V_w \times N_w$$

where  $N_w$  stands for the nitrogen content in a runoff water ( $\text{g l}^{-1}$ );  $V_w$  is an annual total water flow [ $\text{lh}^{-1} \text{y}^{-1}$ ] from soil to the river Prut. River runoff water was sampled within one-week period in March, June and October for each of the study ecosystems using the runoff collector.

Nitrogen contents in a river runoff water and in precipitation water were measured using the following methods:  $\text{NO}_2$  content was measured using the Griess reagent and colorimetric detection (photocolorimeter KФK-3; EuroLab, St Petersburg, Russia);  $\text{NO}_3$  was measured using the Portable Nitrate Sensor (H-401; Ukraine); and  $\text{NH}_4$  was measured using the Nessler's Reagent and colorimetric detection (photocolorimeter KФK-3; EuroLab, St Petersburg, Russia).

#### 2.2.8. N-fixation, uptake and exudation estimates

Our estimated rate of the nitrogen fixation by the black locust accounts in for  $18.8 \text{ g N Tree}^{-1} \text{y}^{-1}$  that is equivalent to  $13.16 \text{ kg N ha}^{-1} \text{y}^{-1}$  due to the tree density of 700 ( $\text{SD} = 81.6$ ) trees  $\text{ha}^{-1}$ . The estimated rate is derived from the measurements within the 2-year experiment on biological  $\text{N}_2$  fixation in field-grown *Robinia pseudoacacia* L. in Austria by Danso et al. (1995). It is assumed based on Danso et al. (1995) results that 21% of the total incoming N accounts for the root-uptake by black locust, which is equivalent approximately to  $3.5 \text{ kg ha}^{-1} \text{y}^{-1}$ . Data of Hangs (2013) on willow uptake efficiency of  $^{15}\text{N}$ -labeled fertilizer (31%) were used to estimate the plant N uptake for the *Sallix* spp. site.

Non-symbiotic N fixation was based on the data for the deciduous forests by Todd et al. (1978) and Swank and Waide (1980) who suggest the estimated rates for the non-symbiotic N fixation in soil of  $8.6 \text{ kg ha}^{-1} \text{y}^{-1}$ , woody litter of  $1.66 \text{ kg ha}^{-1} \text{y}^{-1}$  and leaf litter of  $0.63 \text{ kg ha}^{-1} \text{y}^{-1}$ . The exudation rate of symbiotically fixed nitrogen from the roots of black locust into the soil was estimated using the experimental measurements of Uselman et al. (1999) whose findings show that the *R. pseudoacacia* exude about 2% of the fixed N through their roots as dissolved organic N. For the *Salix* spp. the N-exudates measure was derived from the model balancing.

#### 2.3. Model construction and balancing

Conceptual models for the N cycling for each of the study sites are presented by Fig. 1. The two models differ in the plant N-fixation flow due to the non-fixing trait of *Salix* spp. (Fig. 1). Models were built and analyzed using a simulation and network analysis software for ecological systems EcoNet (Kazanci, 2007). Standing stocks are in  $\text{kg N ha}^{-1}$ , flows are in  $\text{kg N ha}^{-1} \text{y}^{-1}$ .

Initially because of lack of empirical data the network models included some lower quality (literature based) data. Ideally, the missing or lower quality flow rates can be calculated using the steady-state assumption. For the ecosystem model to be at steady state, total flow rate entering and exiting each compartment should

equal each other. However, this model is over determined, meaning that it is not possible to satisfy the steady-state assumption for each compartment by using the degree of freedom allowed by the unknown flow rate values. Therefore some of the flow rates had to be modified slightly to achieve a balanced network model at steady state. However, modifying flow rates that are based on reliable empirical data is not desirable. So we used constrained non-linear multivariable optimization to perturb only the flow rates that were missing empirical data or were based on lower quality data. We assigned confidence ratings for each flow rate, reflecting the quality of empirical data it is based on. We then used the *fmincon* function from Matlab's optimization toolbox, which utilizes the standard Moore-Penrose pseudoinverse approach, to achieve a balanced steady state ecological network model that accurately reflects the collected field data.

#### 2.4. Ecological network analysis

Network system-wide properties were measured at steady state. The assessed indices are fully documented in literature (Buzhdygan et al., 2012), but brief descriptions and abbreviations are listed in Table 1. Turnover time was measured as the N standing stock compared to outflow.

### 3. Results

#### 3.1. Robinia and Salix stand characteristics

Our results on the black locust and willow stem densities, and shoot basal diameter are presented in Table 2. The willow site had higher stem density in comparison to the *Robinia* stands. The *Salix* stands ranged widely in shoot basal diameter; while the black locust site had more even stand characteristics within the study area.

#### 3.2. Nitrogen concentration

The N concentrations in live tissues, litter and soil are reported in Table 3. Live tissue N content was higher in leaves in comparison to the woody parts for both, black locust and willow. *Salix* versus *Robinia* live tissue N content was lower throughout the all compartments. A similar pattern was observed for the litter N content (Table 3) as well as for the fallen leaves (Table 4). Litter N content was near three times higher than in soil for both of the study sites. Runoff water N content was two times higher within the *Robinia* site in comparison to the *Salix* spp. (Table 4). Soil total N content was slightly higher in the black locust site. However, the denitrification rate was higher under the *Salix* stands (Table 4).

#### 3.3. Standing stocks

Dry weight and nitrogen standing stocks are presented in Table 3. Black locust and willow biomass (above-ground dry weight) was  $34.6 \text{ t ha}^{-1}$  and  $13.1 \text{ t ha}^{-1}$  respectively. The total system dry-weight stocks were  $1827.2 \text{ t ha}^{-1}$  ( $6.6 \text{ t N ha}^{-1}$ ) and  $1709.6 \text{ t ha}^{-1}$  ( $5.22 \text{ t N ha}^{-1}$ ) for the *Robinia* and for the *Salix* sites respectively.

Near 91% of the total system N was stored in the soil under the *Robinia* stands. Black locust stored near 5% of the system N in the live tissues, 93% of which was stored in the wood. Less than 1% of the system's nitrogen was in the litter.

Within the *Salix* study site more than 97% of the total system N was stored in the soil, near 1% – in the litter and near 2% – in the live tissues (88% of which was in wood and 12% – in leaves).

**Table 1**  
Description of network system-wide properties.

Network measure, abbreviation	Description	Equation
Total Standing Stock, <b>TSS</b>	The sum of compartmental standing stocks	$TSS = \sum X_i$ , where $X_i$ is the amount of material stored in compartment $i = 1, \dots, n$ , where $n$ is a number of compartments.
Total System Throughflow, <b>TST</b>	The sum of compartment throughflows (total amount of flows within a network)	$TST = \sum T_i$ , where $T_i$ is the total amount of flow through compartment $1, \dots, n$ , where $n$ is a number of compartments. <a href="#">Hannon, 1973</a> ; <a href="#">Finn, 1976</a> ; <a href="#">Han, 1997</a> .
Finn's Cycling Index, <b>FCI</b>	Relative amount of cycling in ecosystem (fraction of total system throughflow that cycle).	$FCI = TST_c / TST$ , $TST_c = C_1 T_1 + \dots + C_n T_n$ , where $TST_c$ is the cycled portion of <b>TST</b> , is the weighted sum of cycling efficiencies $C_i$ of all compartments $i = 1, \dots, n$ , where $n$ is a number of compartments. $C_i = (n_{ii} - 1) / n_{ii}$ , where $n_{ii}$ is the number of times a flow quantity will return to $i$ before being lost from the system, $(n_{ii} - 1)$ is the relative amount cycled. <a href="#">Finn (1976, 1980)</a> .
Relative ascendancy, <b>RA</b> (unitless)	It is system realized development.	$RA = AMI / H_f$ , $AMI = \sum p(T_{ij}) \times \log_2 \{ p(T_{ij}) / p(T_j) \} / p(T_i)$ , $H_f = - \sum (p(T_{ij}) \times \log(p(T_{ij})))$ , where AMI (bits) – the average mutual information, it is the degree of organization with which the exchanges between compartments are processed, $H_f$ is a Shannon ( <a href="#">Shannon, 1948</a> ) flow diversity, $T_{ij}$ is the flow from $i$ to $j$ ; $p(T_{ij})$ is an individual joint probabilities of flows from each species $i$ to each species $j$ given by: $p(T_{ij}) = T_{ij} / TST$ , $p(T_i) = \sum_j p(T_{ij})$ , $p(T_j) = \sum_i p(T_{ij})$ , where <b>TST</b> is a total system throughflow. <a href="#">Ulanowicz, 1986, 1997</a> .

**Table 2**  
Stand characteristics of the black locust and willow study sites of Prut river floodplain in Ukraine.

Stand characteristics	Study site			
	<i>Robinia pseudoacacia</i> L.		<i>Salix</i> spp.	
	Mean $\pm$ SD	Range	Mean $\pm$ SD	Range
Stem Density, trees ha <sup>-1</sup>	700 $\pm$ 81.6 (n = 3)	600–800	1200 $\pm$ 294 (n = 3)	900–1600
Shoot Basal Diameter SBD, cm	12 $\pm$ 3.21 (n = 24) <sup>a</sup>	7–16	5.4 $\pm$ 3.9 (n = 36) <sup>a</sup>	0.5–17.7

<sup>a</sup> SBD was measured at 10 cm and 15 cm above-ground level for stems of *Robinia* and *Salix* respectively.

### 3.4. Litterfall and decomposition

Decomposition rate of black locust litter was 0.4 and litter decomposition respectively was 1.9 t ha<sup>-1</sup> y<sup>-1</sup> (dry weight) and 21.7 kgN ha<sup>-1</sup> y<sup>-1</sup>. Total litterfall (leaves and woody parts) was

2 t ha<sup>-1</sup> y<sup>-1</sup> (dry weight) and 31.29 kgN ha<sup>-1</sup> y<sup>-1</sup> within the *Robinia* study site.

The *Salix* litter decomposition rate (0.29) was considerably lower than under the *Robinia* stands. Therefore, litter decomposition under the *Salix* spp. was two times lower (1.06 t ha<sup>-1</sup> y<sup>-1</sup> (dry

**Table 3**  
Dry weight, nitrogen concentration and nitrogen standing stocks in black locust and willow study sites of Prut river floodplain in Ukraine.

Network compartment	Dry weight (kg h <sup>-1</sup> )		Nitrogen concentration (% of dry weight)		N standing stocks kgN h <sup>-1</sup>
	Mean $\pm$ SD, n = 3	CV	Mean $\pm$ SD, n = 3	CV	
<b><i>Robinia pseudoacacia</i> L.</b>					
Leaves	1487.25 $\pm$ 273.2	0.18	3.22 $\pm$ 0.30	0.09	47.86
Branches	6020.08 $\pm$ 1178.1	0.20			50.63
trunk	27,117.85 $\pm$ 5579.8	0.21	0.84 $\pm$ 0.10	0.12	228.06
Wood	33,137.92 $\pm$ 6757.9	0.2			278.69
Litter	4732.8 $\pm$ 761.8	0.16	1.15 $\pm$ 0.70	0.61	54.29
Soil	1,754,666.67 $\pm$ 1548.1	0.001	0.34 $\pm$ 0.08	0.24	5965.87
<b><i>Salix</i> spp.</b>					
Leaves	855.7 $\pm$ 325.1849	0.38	1.43 $\pm$ 0.196	0.14	12.2365
Wood	12,279.35 $\pm$ 4925.873	0.40	0.76 $\pm$ 0.09	0.12	93.32307
Litter	3664.75 $\pm$ 758.295	0.21	0.97 $\pm$ 0.34	0.35	25.85
Soil	1,692,800 $\pm$ 215,457.9	0.13	0.3 $\pm$ 0.07	0.23	5078.4

**Table 4**

Measured flow rates and nitrogen concentrations for parametrization of flows in black locust and willow study sites of Prut river floodplain in Ukraine.

Study site	<i>Robinia pseudoacacia</i> L.		<i>Salix</i> spp.	
	Mean $\pm$ SD, n = 3	CV	Mean $\pm$ SD, n = 3	CV
<b>Flow rates</b>				
Decomposition rate	0.4 $\pm$ 0.21	0.53	0.29 $\pm$ 0.05	0.17
Denitrification, kg N ha <sup>-1</sup> y <sup>-1</sup>	0.52 $\pm$ 0.003	0.01	0.61 $\pm$ 0.067	0.11
Falling woody parts, kg dry weight ha <sup>-1</sup> y <sup>-1</sup>	532.3 $\pm$ 97.3	0.18	188.2 $\pm$ 72.3	0.38
<b>Nitrogen concentration</b>				
Precipitation water, mg N l <sup>-1</sup>	0.48 $\pm$ 0.11	0.23	0.48 $\pm$ 0.11	0.23
Runoff water, mg N l <sup>-1</sup>	11.3 $\pm$ 1.45	0.13	5.3 $\pm$ 0.88	0.17
Falling leaves, % of dry weight	1.8 $\pm$ 0.16	0.09	1.01 $\pm$ 0.04	0.04

**Table 5**

Soil properties (1–20 cm depth) under the black locust and willow stands in Prut river floodplain in Ukraine.

Soil properties	Study site			
	<i>Robinia pseudoacacia</i> L.		<i>Salix</i> spp.	
	Mean $\pm$ SD, n = 3	CV	Mean $\pm$ SD, n = 3	CV
Bulk density (g cm <sup>-3</sup> )	0.88 $\pm$ 0.001	0.001	0.89 $\pm$ 0.03	0.03
Organic Carbon (%)	5.58 $\pm$ 0.24	0.04	5.44 $\pm$ 0.26	0.05
Soil pH (KCl)	4.7 $\pm$ 0.9	0.2	5.3 $\pm$ 0.15	0.03
C/N ratio	16.4 $\pm$ 3.1	0.19	18.6 $\pm$ 3.92	0.21

**Table 6**

Flow matrices (from column to row oriented) at system steady state of nitrogen models of the black locust and willow study sites of Prut river floodplain in Ukraine.

Flows, kg N ha <sup>-1</sup> y <sup>-1</sup>						
Study site	<i>Robinia pseudoacacia</i> L.					
	Soil (f)	Wood (f)	Leaves (f)	Litter (f)	Environment (input (z))	Environment (output (y))
Soil	0	1.11765	0	25.3271	14.995	38.7957
Wood	2.64379	0	22.2377	0	23.801	0
Leaves	0	46.6675	0	0	0	0
Litter	0	0.897398	24.4297	0	1.39E–08	0
Study site	<i>Salix</i> spp.					
Network compartment	Soil (f)	Wood (f)	Leaves (f)	Litter (f)	Environment (input (z))	Environment (output (y))
Soil	0	7.73629e–07	0	10.5419	17.246	18.4078
Wood	9.37986	0	3.5935	0	0	0
Leaves	0	11.89	0	0	0	0
Litter	0	1.08336	8.29649	0	1.162	0

weight) and 10.3 kgN ha<sup>-1</sup> y<sup>-1</sup>) than that under the black locust trees. Total litterfall of the willow trees was considerably lower (1.04 t ha<sup>-1</sup> y<sup>-1</sup> (10.1 kgN ha<sup>-1</sup> y<sup>-1</sup>)) than that of the black locust.

### 3.5. Soil properties

Both of the study sites have the gray-forest soils. Soil properties for each of the study sites are listed in Table 5. Soil organic carbon and bulk density were of the similar value ranges for the both sites. However, the soil pH and C/N ratio were lower under the *Robinia* stands.

### 3.6. Nitrogen-budget models and system-wide properties

Nitrogen-cycle networks and the input parameters for the models are presented in Fig. 1. The steady-state flow matrices are given in Table 6 and system-wide properties are reported in Table 7.

Both, total standing stock (TSS) and throughflow (TST) were considerably higher under the black locust (TSS = 6.35 t N ha<sup>-1</sup>, TST = 0.16 t N ha<sup>-1</sup> y<sup>-1</sup>) than that under the willow trees (TSS = 5.2 t N ha<sup>-1</sup>, TST = 0.06 t N ha<sup>-1</sup> y<sup>-1</sup>). Cycling index was 10% higher in the willow ecosystem (FCI = 0.41) than that in the

black locust site (FCI = 0.31). Relative ascendancy values are near the same for both study systems (Table 7).

Turnover time of N in the system was considerably higher under the *Salix* trees in comparison to the *Robinia* site of the floodplain (Table 7). Soil N pool demonstrated the highest turnover time

**Table 7**

System-wide properties of nitrogen networks of black locust and willow study sites of Prut river floodplain in Ukraine.

System-wide properties	<i>Robinia pseudoacacia</i> L.	<i>Salix</i> spp.
Total Standing Stock TSS, kg N ha <sup>-1</sup>	6346.71	5209.81
Total System Throughflow TST, kg N ha <sup>-1</sup> y <sup>-1</sup>	162.117	63.1928
Finn's Cycling Index FCI, unitless	0.31454	0.407136
Relative ascendancy RA, unitless	0.61	0.58
Turnover time, y		
Network compartment	<i>Robinia pseudoacacia</i> L.	<i>Salix</i> spp.
Soil	143.97	182.76
Wood	5.72	7.19
Leaves	1.03	1.03
Litter	2.14	3.37
Total System property	164	283

relative to the other compartments for both study sites. N residence time in the litter pool was 2–3 times higher than in live tissues in both study ecosystems.

#### 4. Discussion

Most of the black locust studies are focused on the areas cultivated with *Robinia* (Boring and Swank, 1984; Berthold, 2005; Tateno et al., 2007) and few investigate the naturally invaded ecosystems (Akamatsu et al., 2011). Current study focuses on the floodplain naturally colonized by black locust and compares the nitrogen budgets under the black locust and under the indigenous willow stands.

Stem density of black locust (700 trees ha<sup>-1</sup>) was near ten times lower than usually reported by the studies of cultivated black locust. As a result both, plant biomass standing stock as well as plant N standing stock were lower in our study. But our data are consistent with the study of the naturally invaded floodplains by Akamatsu et al. (2011) who reported the natural colonization of Chikuma River riparian area with the density of 602 black locust stems ha<sup>-1</sup>.

Our findings show that live tissue nitrogen content was higher in black locust in comparison to the willow trees. This is consistent with the findings reported in the literature (Boring and Swank, 1984; Parolin et al., 2002; Akamatsu et al., 2011). Tateno et al. (2007) showed that an annual N input through the litterfall under black locust is higher than under N non-fixing trees. Our data are consistent with these findings and report 2.7 times higher litterfall by black locust than that by willow trees.

Some authors suggest that the black locust litter decomposes slowly due to the dominance of lignin (Uselman et al., 1999). Alternative studies report that black locust litter decomposes faster than that of non-fixing trees (Tateno et al., 2007; Malcolm et al., 2008). Decomposition rate of black locust litter at our study was 0.4, which is 10% higher than it is observed in case of the willow ecosystem (0.29). Moreover, the turnover time of N in the litter pool under the willow trees was 1.23y higher relative to the black locust site. The slow decomposition rate of *Salix* spp. litter could be explained by high content of lignin that makes cellulose more resistant to degradation. Also number of studies found slower decomposition rates for the species with initially low N content in contrast to the faster decomposition of high N content litter (Aziz et al., 2011). Thus, slower decomposition rate of willow litter could be attributed to the lower initial N content in comparison to the black locust litter.

Our results show that less than 1% of the total system N stock is stored in the litter within both of the study sites. Furthermore, N turnover time in the litter pool is the lowest in comparison to the other compartments in both of the study ecosystems. It could be associated with the flooding regime. Study floodplain inundates annually for 2–3 months (March–April) during which the water removes litter from the surface. This could attribute to the low amount of surface litter standing stock. Also it is shown by Kitamura (2012) that in deciduous leaves, most leaching occurs within a few days of immersion in water. Up to 25% of the initial leaf dry weight of *Salix* spp. can be lost by leaching in the first 24 h of immersion (Webster and Benfield, 1986).

It has been reported the higher rates of N accumulation by soil under the black locust stems than that under the other trees (Boring and Swank, 1984; Tateno et al., 2007; Malcolm et al., 2008). It is consistent with our results. We found higher N soil standing stock and considerably lower turnover time for the soil N pool under the *Robinia* than under the *Salix* spp. The soil N concentration was also slightly higher for the black locust site (0.34%) than that for the willow ecosystem (0.3%).

Total nitrogen storage (standing stock and throughflow) is 20% higher in the black locust network than in the willow ecosystem.

Number of studies suggests that black locust is disadvantageous for the soil development (Tateno et al., 2007) because of the N-enrichment can lead to soil degradation through the strong nitrification, leaching and acidification (Berthold, 2005). Study soil under the black locust was more acidic than that under the willow stands. Soil C/N ratio was higher in the willow study site that might be explained by slower decomposition rates within this system in comparison to the black locust.

In total our data report higher N-flow rates and faster N-cycling processes in the *Robinia* ecosystem relative to the *Salix* site of the study floodplain. This attributes to the almost three times higher values of the total system N throughflow (TST) within the black locust site than within the willow part of the floodplain, resulting also in two times higher N outputs to the river. Also the cycling of nitrogen (fraction of TST that is circulating) was higher in *Salix* (40%) than in the *Robinia* ecosystem (30%). This indicates that the use (conservation) of N inflows within the willow ecosystem is more efficient than that in the black locust. Similar results derive from the ratio of TST to the total system outflow. It is higher within the black locust (4.2) in comparison to the willow site (3.4). Turnover time data indicate that total ecosystem N-related processes are 2 times faster in the *Robinia* site of the floodplain in comparison to the *Salix* spp. Our network data on relative ascendancy show that both ecosystems are on the same stage of development (Ulanowicz, 1986, 1997), namely 60% of their development capacity. Further system development (up to 40% of the system development capacity) in the combination with the rapid and non-conservative use of N (with high outflow fraction to the river (99% of the total system outflow)) might result in subsequent significant increase of N runoff into the river in the black locust site.

#### 5. Conclusions

In this study we compare the nitrogen budgets of two sites of floodplain ecosystem, one is colonized by invasive black locust and the other is forested by the indigenous *Salix* spp.

Our data report the considerably higher N-flow rates, storage and the faster total N-related ecosystem processes in the invaded *Robinia* site in comparison to the indigenous *Salix* site of the study floodplain. Black locust affects the nitrogen cycle in the study riparian ecosystem through the N-fixation, high N-content litterfall and rapid litter decomposition that results in the increase of the nitrogen runoff into the river and contaminates the water.

Black locust appeared to reduce the conservation values of the colonized areas of riparian ecosystem and adjacent river Prut. Restoration strategies that remove the black locust from invaded sites and prevent the further invasion will slow down the contamination. However, high availability of nitrogen in soil and soil degradation might have a long-term effect on floodplain ecosystem after the restoration.

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