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Examination of the decomposition of willow (*Salix sp.)*, poplar (*Populus sp.*), reed (*Phragmites australis*) and mixed leaf litter with litterbag technique

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SUMMARY

Leaf litter decomposition is one of the main ecological material cycle processes in waterfront areas. In this microcosm experiment, the rate of decomposition of the most frequently occurring dominant waterside plants were examined in the summer months of 2022 in a class "A" evaporation pan, using litterbag technique. The study provides information about the decomposition dynamics of willow (Salix sp.), poplar (Populus sp.), reed (Phragmites australis) and different leaf litter mixture combinations. Dry mass, exponential decay coefficient and the chemical parameters of the water (pH, conductivity, NH_4^+, PO_4^{3-}, SO_4^{2-}) were determined during the 84 days long experimental period. The weight loss curves showed negative exponential pattern in every case. On average, the different samples lost ~ 57% of their initial dry mass during the experimental period. The largest mass loss was measured in case of poplar (67.2%), while reed leaves had the smallest mass loss (47.25%). Based on the results, it cannot be proven, that mixed leaf litter accelerates the rate of decomposition.

Keywords: *leaf litter decomposition; litterbag technique; willow; poplar; reed*

INTRODUCTION

Biomass decomposed by aquatic saprotrophic organisms plays an important role in the regeneration of soluble substances and contributes to the cycle of carbon, nitrogen and phosphorus in freshwater biotopes (Bärlocher and Kendrick, 1981). Next to the autochtonous components, allochtonous sources [especially leaves (>60%)] provide a large amount of nutrients and energy (Dobson and Frid, 1998). The speed of leaf litter decomposition depends on the one hand on biotic factors (e.g. saprotrophic organisms, type of leaf litter and lignocellulose content, depending on the age of leaves) and on the other hand on abiotic factors (e.g. temperature, water movement and chemical parameters of the water) (Chen et al., 2019). The mix-up of different types of leaf litter can also have an effect on the rate of decomposition and the dynamics of nutrient release (Lecerf et al., 2011).

In the floodplain of rivers, shallow lakes and highland and lowland streams, the most typical domestic tree species are willows (*Salix sp.*) and poplars (*Populus sp.*) (Bagi et al., 1996). In addition, the common reed (*Phragmites australis*) is the dominant herbaceous plant species of the coastal zone of Hungarian waters.

Determining the leaf litter decomposition rates can help to better understand the material cycle processes of aquatic ecosystems.

The main aim of the study was to evaluate the decomposition rate of willow (*Salix sp.*), poplar (*Populus sp.*), reed (*Phragmites australis*) and different leaf litter mixture combinations (mixed in equal proportions) in a standard class „A" evaporation pan. The decomposition rates were studied in the presence of macroinvertebrates, so litterbags with 3 mm mesh size were used. Hypothesis that mixed leaf litter increases the rate of decomposition was tested. An additional purpose was to draw conclusions from the changes in water chemical parameters.

MATERIALS AND METHODS

The research was launched at the Agrometeorological Research Station of Hungarian University of Agriculture and Life Sciences, Keszthely (N: 46° 44' 7,93", E: 17° 14' 16,65", 143 m) on June 10. 2022.

The experiment was carried out in a standard class „A" evaporation pan because it is suitable for conducting aquatic microcosm experiments. It's size and arrangement fitted the legal requirements of WMO (outside and inside white, 120 cm diameter, 25 cm deep, filled with tap water, layed on double wooden lattice) (Gombos, 2011). Its bottom was filled with sediment from lake Balaton in 3 cm layer to provide micro- and macroorganisms. By using the evaporation pan, the negative effect of drifting was eliminated.

Leaf litter decomposition of pre-dried willow, poplar, reed and mixed leaf litter were examined using litterbag technique, following the method of Bärlocher et al. (2005), between 10. June 2022 and 2. September 2022. 126 litterbags were made in a size of 15x15 cm out of non-biodegradable material with a mesh size of 3 mm. In order to ensure the access of macrofauna to the substrate, the mesh size of the litterbags must be at least 2 mm (Harmon et al., 1999). To stimulate autumn circumstances, the leaf litter samples were dried beforehand. Each bag was filled with 10 g of willow, poplar or reed leaves or willow-poplar, reed-willow, reed-poplar leaf mixtures, mixed in 1:1 ratio or willow-poplar-reed leaves, mixed in 1:1:1 ratio. For each of the 7 variations, 6 litterbags were made, with 3 repetitions. The mouth of the filled bags was sealed, then they were tied to modified plastic baskets and were weighted to the bottom of the evaporation pan with stones to avoid replacement.

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A total of 6 sampling happened on the 14th, 28th, 42nd, 56th, 70th and 84th days after the placement. During each sampling 3 of each type of samples were processed, 21 litterbags per occasion. The litterbags were taken to the laboratory and were unwrapped and depurated from contaminations, macroinvertebrates and other foreign materials, using forceps, laboratory sieve and tap water. After that, they were placed on metal laboratory trays and dried until their weight was constant (usually for 14 days). Then the dry weights were remeasured using digital analytical scales.

To describe leaf litter decomposition, the exponential decay model was used which was first described by Jenny et al. (1949). The rate of decomposition was determined using the following equation:

$$M_t=M_0*e^{-kt} \tag{1}$$

where „M_t" (g) is the mass at time, „M_0" (g) is the mass at time 0, „k" (day^{-1}) is the exponential decay coefficient and „t" (day) is time. Expressing the exponential decay coefficient from the equation, the rate of decay of each plant species can be categorized as „fast" where $k > 0.01$, „medium" where $k = 0.005$-0.01 or „slow" where $k < 0.005$ (Bärlocher et al., 2005; Petersen and Cummins, 1974).).

The calculation of halving times was based on the following formula of Bärlocher et al. (2005):

$$T_H= \ln 2*k^{-1} \tag{2}$$

where „T_H" (day) is halving time and „k" is the exponential decay coefficient.

The samplings of the water body, where leaf litter decomposition took place happened just before the placing of the litterbags, furthermore always before litterbag samplings, so 7 samples were processed in total. To determine the pH and conductivity of the water samples, fast digital measuring devices (Adwa AD110 pH and thermometer and Adwa AD310 conductivity-temp portable meter) were used. The NH_4^+, PO_4^{3-} and SO_4^{2-} content of the samples were determined by spectrophotometric method (using Lovibond Multidirect Spectrophotometer). The water samples were processed on September 6. 2022. Until this date, the samples were stored in a deep freezer to preserve their chemical quality.

RESULTS AND DISCUSSION

The pH of the water was mildly alkaline (8.1 ± 0.6) during the experimental period, which was suitable for most of the macro- and microinvertebrates.

Only NH_4^+ and PO_4^{3-} (*Table 1*) showed greater variability. The main reason of the high standard deviation values was the rapid leaching at the beginning (first 24 hours) of the experiment. In their study Simon et al. (2021) have also observed the initial jump-like mass loss and NH_4^+, PO_4^{3-} leaching from their *Solidago virgaurea* samples in Hévíz Lake and Hévíz canal. It is also possible, that the variability of the NH_4^+ -values was partially caused by the activity of nitrifying and denitrifying organisms, which entered the evaporation pan with the sediment, leaf litter or by the wind. Additional slight variabilities may have also been caused by the diluting effect of natural phenomena with higher precipitation rates or by the daily methodical refilling of the evaporation pan. Temperature extremes may also have caused occasional changes and variabilities in the water chemical parameters through the destruction of microbes. Too high NH_4^+ and PO_4^{3-} concentrations are unfavorable, because they cause eutrophication, which may lead to the disappearance of microorganisms with narrower tolerance range (Jespersen et al., 2016).

The other variables did not differ to a greater extent. SO_4^{2-} concentration was almost constant during the experimental period. It was unceasingly below the permissible concentration limit of 250 mg L^{-1} for drinking water, which was defined by the Hungarian 201/2001 (X. 25.) Government decree.

After an initial jump-like rise, conductivity values increased almost monotonously. In his research, Garbowsky (2019) described that due to the dissolving substances, the electrical conductivity of the water increases parallel to the residence time of the decomposing substances in the water. Hasanuzzaman and Mahmood (2015) have also verified the former statement. They described that, there is a direct proportionality between the mass loss of the leaf litter and the amount of dissolved material in the water and the conductivity value.

Table 1: **Water quality parameters during the experimental period**

pH	8.1 ± 0.6
Conductivity [µS cm^{-1}]	740.6 ± 85.9
NH_4^+ [mg L^{-1}]	0.22 ± 0.19
PO_4^{3-} [mg L^{-1}]	0.35 ± 0.3
SO_4^{2-} [mg L^{-1}]	70.2 ± 23.7

The change in mass of each leaf litter types can be seen on *Figure 1*. The initial jump-like mass loss can be attributed to leaching in the first phase of decay. On average, the different samples lost ~ 57% of their initial mass during the 84 days of the experiment. The largest mass loss was measured in case of poplar (67.2%), while reed leaves had the smallest mass loss (47.25%).

The fit of the exponential curve was the most optimal in case of reed-poplar (R^2=0.9688), while the value was the lowest in case of willow (R^2=0.9424).

Figure 2 illustrates the values of the exponential decay coefficient (k) in our samples. Based on the k-values poplar (0.00685 ± 0.00082), willow-poplar (0.00587 ± 0.00055) and reed-poplar (0.00527 ± 0.00078) samples were categorized into „medium" ($k = 0.005$-0.01) decomposition category, while willow (0.00465 ± 0.00069), reed (0.00413 ± 0.00042), reed-willow (0.00458 ± 0.00070) and willow-poplar-reed (0.00484 ± 0.00069) samples fell into „slow" ($k < 0.005$) decomposition category.

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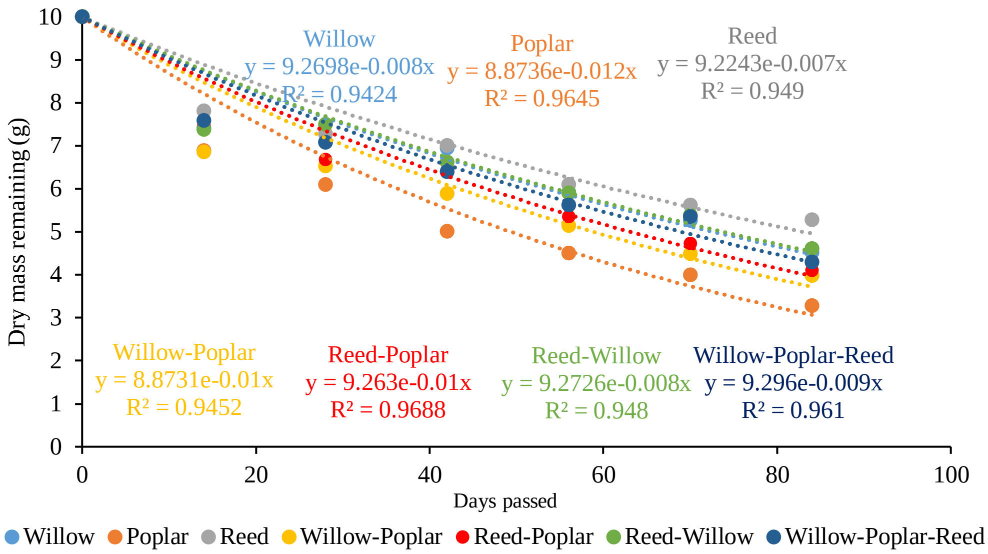
Figure 1: **Changes in mass over time**

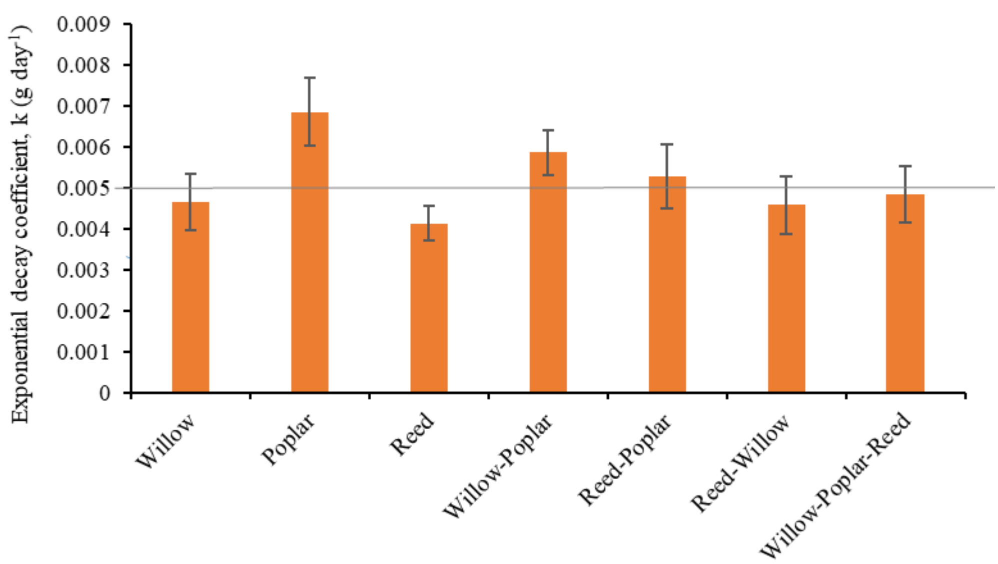


Based on the results of the One-Way ANOVA (at a significance level of 0.05) a significant difference can be observed between the k-values of willow and poplar ($p < 0.05$), poplar and reed ($p < 0.05$), poplar and reed-poplar ($p < 0.05$), poplar and reed-willow ($p < 0.05$), poplar and willow-poplar-reed ($p < 0.05$), reed and willow-poplar ($p < 0.05$), willow-poplar and reed-willow ($p = 0.03$) samples. No significant differences were observed in case of the other variations.

The differences between the decomposition indicators of each type of samples can be traced back to the morphological, physical and chemical properties (cellulose, lignin content and C:N ratio) of the leaves of the different plant species. A high N and P concentration compared to the C content of the leaves, as well as the low concentration of complex polymers, that are difficult to degrade (e.g. lignin) result in fast decomposition, while low concentrations of N and P cause slower leaf litter decomposition processes (Kadlec and Wallace, 2009). While the former can be observed in the case of willow and poplar (Ágoston-Szabó et al., 2016), the latter is more typical for reed (Ágoston-Szabó and Dinka, 2008).

The type and substrate specificity of the degrading micro- and macroorganisms could also largely determine the rates of leaf litter decomposition.

Figure 2: **Distribution of exponential decay coefficient in different litters**



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Knowing the k-values, the halving times (*Table 2*) were also determined. Based on the results of the research, the statement of Chapman and Koch (2007), that the co-presence of different leaf litter types tend to accelerate the process of decomposition and that a more homogenous substrate creates better conditions for the adaptation of saprotrophic organisms could not be confirmed. The decomposition time of the mixed samples was an almost exact average of the decomposition time of the 2 or 3 leaf species which they consisted of.

In their research, Simon et al. (2019) investigated the leaf litter decomposition of willow, poplar and mixed leaf litter. They were also unable to demonstrate the significant stimulating effect of mixed leaf litter on decomposition.

Table 2: **Halving times of willow, poplar, reed and mixed leaf litter samples**

Leaf litter type	Halving time (day)
Willow	151.8 ± 22.9
Poplar	102.5 ± 12.8
Reed	169.3 ± 17.3
Willow-Poplar	119.1 ± 11.5
Reed-Poplar	133.9 ± 19.4
Reed-Willow	154.6 ± 26.5
Willow-Poplar-Reed	145.4 ± 19.4

CONCLUSIONS

Comparing our data to the results of further researches on the topic, different or even significantly different decomposition rates can be found in case of the plant species examined in this article. Possible differences are partly due to the timing and location of the study sites (Asaeda and Nam, 2002).

In this microcosm experiment, decomposition rates were monitored in an isolated pool in the absence of drifting, therefore the results should primarily be compared with the results of still water experiments, where drifting does not play a major role in mass loss.

The rate of decomposition of plant material is also highly influenced by environmental conditions such as temperature (Bärlocher et al., 2005). In her research Hubai (2017) confirms, that a significant relationship can be observed between the speed of decomposition and temperature. Setting up the experiment in spring, as opposed to winter, ensures faster mass loss (Wrubleski et al., 1997). Migliorini and Romeo (2020) predicted, that global warming might accelerate decomposition rates in aquatic ecosystems, so it would also be worthwhile to examine the relationship between this ecological process and climate change.

Leaf litter decomposition is a complicated multifactorial process, which is largely determined by the local flow conditions, water quality, climatic-, microbiological- and human factors. Therefore, in order to map the leaf litter decomposition dynamics of various water bodies, consecutive site-specific investigations would be necessary.

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