Linking microbial enzymatic activity to CO₂ outgassing from floodplain backwaters under variable hydrological connectivity.

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Introduction

River-floodplain systems play an important role in dissolved organic matter (DOM) cycling and carbon mineralization. Here, the driving force for lateral exchange processes is hydrological connectivity, which influences numerous exchange processes between backwaters and the main river. Surprisingly, the role of floodplains, which are extending the residence time of organic molecules in transport, is poorly studied regarding the processing of DOM by bacteria. When faced with diverse DOM sources, bacteria produce a broad array of enzymes (Chróst 1991). The measurements of extracellular enzymatic activity (EEA), both free-living and associated with particles allow to examine carbon sources and DOM utilization patterns by microorganisms (Foreman et al. 1998; Romaní et al. 2006). This approach helps understanding the partitioning of DOM-substrate, either for bacterial growth or respiration. For lakes it has been suggested that the in-lake respiration is the main process that affects the partial pressure of CO2 (pCO2), and thus the carbon flux. The role of flood plains in carbon flux (CO₂) to the atmosphere remains largely unknown. High CO₂ concentrations in surface waters originate largely from *in situ* respiration of organic carbon (see e.g. Mayorga et al. 2005). However, due to photosynthetic uptake of CO₂, under-saturated conditions may occur at times. Thus, surface waters may serve as either sinks or sources of CO₂.

Hyphotheses

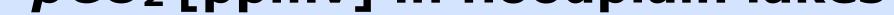
1. Retention time and the degree of connectivity and water exchange is an important driving factor for C-remineralization. The existence of floodplains and their hydrological connectivity with the main stem impacts the overall carbon flux (CO₂).

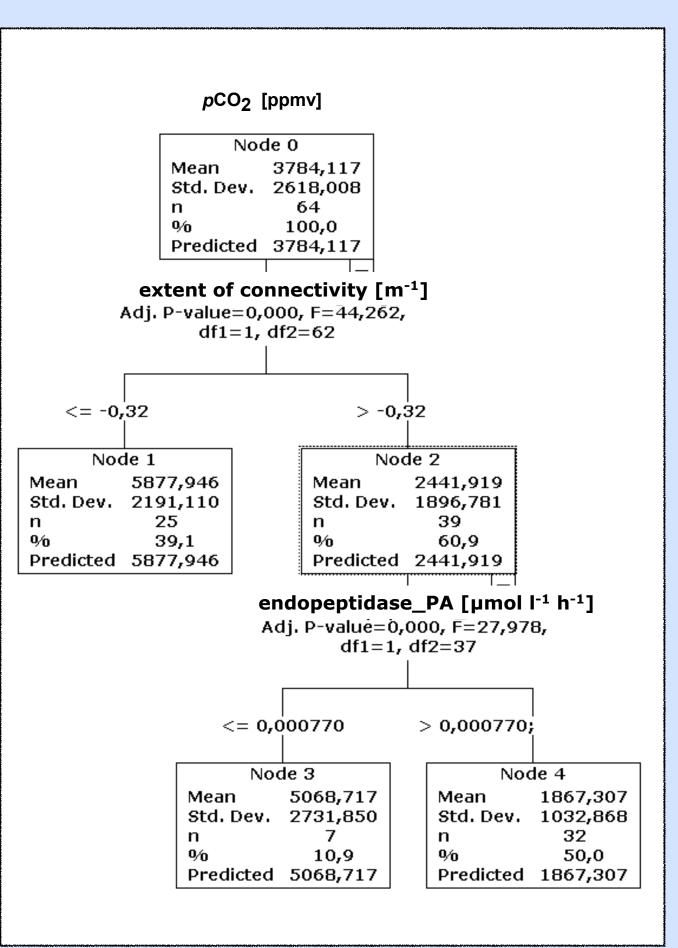
2. The degree of connectivity in "active" river-floodplain systems affects substrate partitioning, mirrored by extracellular enzymatic activities (EEA) both associated with particles (PA) and in the ambient water (FL).

Results and discussion

*p*CO₂ [ppmv] in floodplain lakes

Floodplain lakes as sources of CO₂



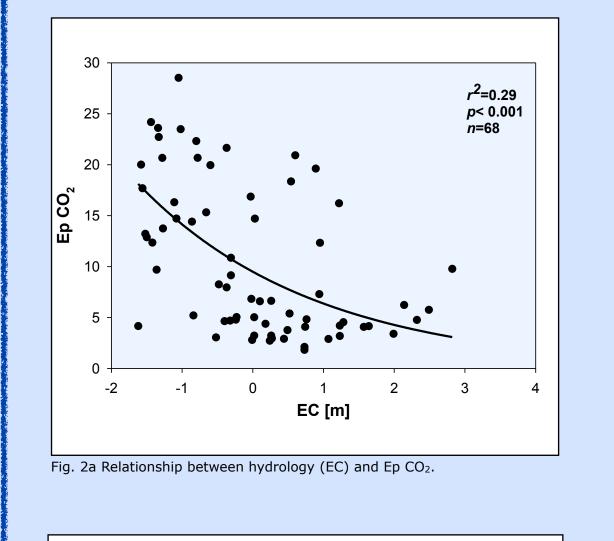


Search Classification tree analysis was applied to predict responses on *p*CO₂ based on set of independent variables (fig. 1). The Classification tree revealed that the most important variable which influenced pCO_2 in this river-floodplain system was hydrological connectivity (EC). During connected conditions, lower *p*CO₂ was noted. When floodplain lakes were connected pCO_2 was also strongly affected by the activity of endopeptidase (N-acquiring enzyme) associated with particles.

Fig. 1 Classification Tree for pCO2 in river-flooplain system. Number of Nodes=5, number of terminal Nodes=3. Independent variables: Slope ratio, Fluorescence index ratio, SUVA₂₅₄ [I mg⁻¹ m⁻¹], chl *a* [µg l⁻¹], EC [m⁻¹], heteroptrohic respiration [µg l⁻¹ h⁻¹], BSP in ambient water and associated with particles [µgC l⁻¹ h⁻¹] activity of N-, C-acquiring enzymes and phox in ambient water and N-, C-acquiring enzymes and phox associated with particles [µmol l⁻¹ h⁻¹]

Importance of particle associated (PA) and ambient water

The importance of variable hydrological connectivity for CO₂ outgassing tendency in different subsystems of the river-floodplain.



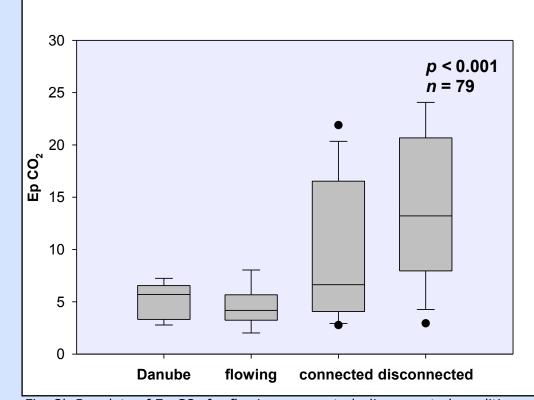


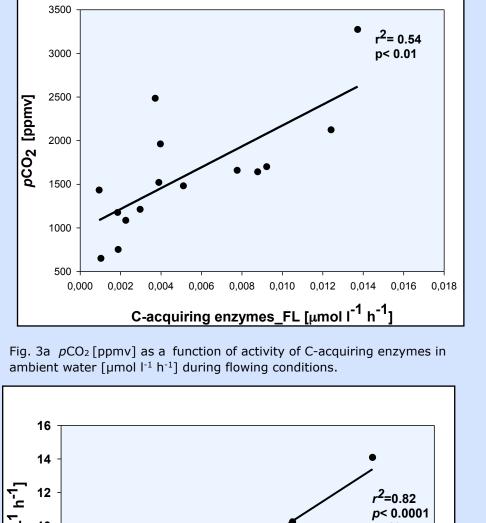
Fig. 2b Boxplots of Ep CO₂ for flowing, connected, disconnected conditions and mai channel of the Danube. The boundaries of the box plot indicate the 25th and 75th pints indicate outliers, the solid line in the box marks

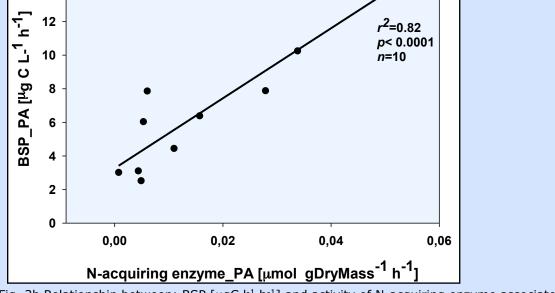
Along with a gradient of connectivity, excess partial pressure (Ep) of CO₂ decreased with increasing connectivity to the main channel (fig. 2a).

Ep of CO₂ during disconnected conditions was significantly higher than during connected conditions. When floodplain lakes were disconnected, **3** times higher values of Ep CO₂ were noted (fig. 2b).

The magnitude of CO₂ outgassing tendency in floodplain lakes depends on the type of hydrological connectivity.

enzymes for *p*CO₂ during flowing conditions





vith particles [gDry mass l⁻¹ h⁻¹] during flowing conditions

ANOVA ^{a,b}							
	Model		Sum of Squares	df	Mean Square	F	Sig.
		Regression	4821188,020	3	1607062,673	12,261	,001 [°]
	1	Residual	1441776,015	11	131070,547		
L		Total	6262964,036	14			

a dependent variable: pCO_2 ; b predictors: Slope ratio, fluorescence index ratio, activity of

 \blacktriangleright Linking EEA to pCO_2 indicated which part of metabolized DOM pool contributed into pCO_2 or rather supported bacterial growth in floodplain lakes.

> Sectivity of C-acquiring enzymes in ambient water was positively linked to pCO_2 (fig.3a). This implied that, at least in hydrologically dynamic stations, increased pCO_2 was largely driven by degradation of carbohydrates. Strong relationship of N-acquiring enzyme with bacterial secondary production (BSP) confirmed that the EEA associated with particles was important and supported bacterial growth in this system (fig 3b).

The multiple regression model revealed that during flowing conditions the best predictors for pCO_2 were Slope ratio, fluorescence index and activity of Cacquiring enzymes in ambient water (Adj $r^2=0.71$, p<0.01, tab. 1).

Conclusions

1. The hydrological connectivity of floodplain lakes with the main channel is the most important factor that drives *p*CO2 in the river-floodplain system.

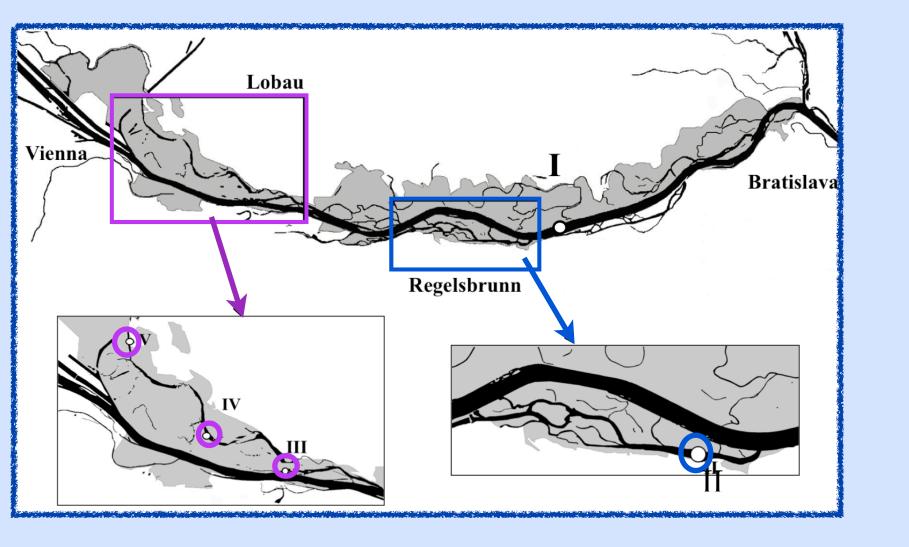
2. Floodplain lakes are supersaturated with CO₂, which indicates that they serve as CO₂ sources. The Ep of CO₂ in floodplain lakes depends on the degree of hydrological connectivity. Severing the connection between river and its floodplain will have consequence for CO₂ flux: the tendency for outgassing of CO₂ is stronger with increased disconnection of floodplain sections from the main channel.

3. Existence of active floodplains and establishment of flooding conditions is important for the fate of enzymatically processed DOM.

In dynamic river-floodplain systems the activity of enzymes associated with particles is mainly responsible for supporting bacterial secondary production. However, C-acquiring enzymes in ambient water play an important role in utilization of allochthonous, high molecular weight DOM, which strongly contributes to pCO_2 during flooding conditions.

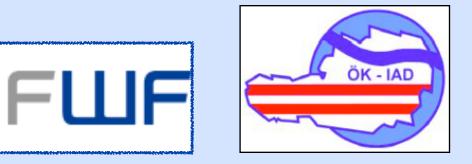
Study site

Sampling locations in the Danube river-floodplain near Vienna (Austria) which represent a gradient of connectivity to the main river.



Acknowledgements

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Methods

Bacterial parameters

- Sector Extracellular enzymatic activity (EEA)
- C-acquiring enzymes:-β-d-glucosidase, a-d-glucosidase, β-d-xylosidase, cellobiohydrolase
- •N-acquiring enzyme:endopeptidase (Hoppe, 1993)
- phenol oxidase (phOx) (Pind et al. 1994)
- Bacterial secondary production (BSP) (Fuhrman and Azam, 1982)

 \subseteq Extent of connection $[m^{-1}]$ (Peduzzi et al. 2008) DOM quantity and quality •DOC [mg l⁻¹] (Benner and Strom 1993) •DOM origin: FI ratio (McKnight et al. 2001) • Molecular size indicator: Slope ratio (Helms et. al, 2008) •Aromaticity: SUVA₂₅₄ (Weishaar et al. 2003) Chlorophyll *a* (Lorenzen, 1967)

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