ENZYME ACTIVITIES WITHIN A COMMUNAL WASTEWATER TREATMENT PLANT: TRENDS AND CORRELATIONS

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Introduction

Enzyme activities are suspected to play a key role in the degradation process of organic wastewater micropollutants. The aim of this work was to monitor their activities over a prolonged period and to determine intercorrelations between enzyme activities as well as dependencies on process and wastewater parameters.

Therefore, the activity of seven hydrolases in activated sludge taken from the main communal wastewater treatment plant (WWTP) of Trier (Germany), were assayed. The monitored enzymes were esterase (EST), phosphatase (PME), phosphodiesterase (PDE), α-glucosidase (α-GLU), β-glucosidase (β-GLU), sulfatase (SUL) and L-alanine aminopeptidase (LAA).

WWTP Trier

- Population equivalent: 170,000; utilization capacity: ~85%
- Wastewater sources: mainly domestic wastewater; industry (3 sparkling wine producers, 1 tobacco factory, 1 rolling mill).

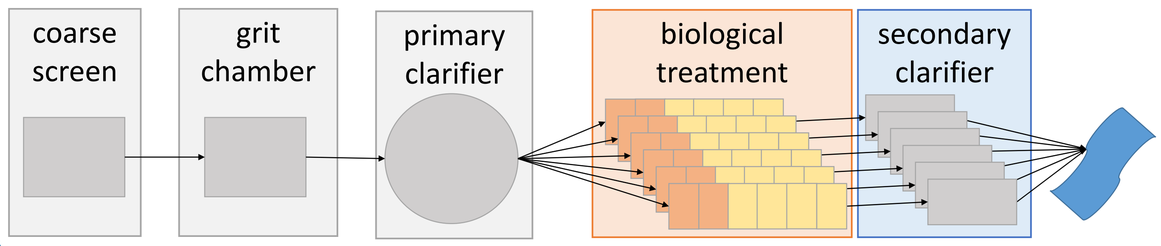


Fig. 1 Layout of the main communal WWTP of the city of Trier, Germany.

Materials & Methods

Essential monitored wastewater and process parameters:

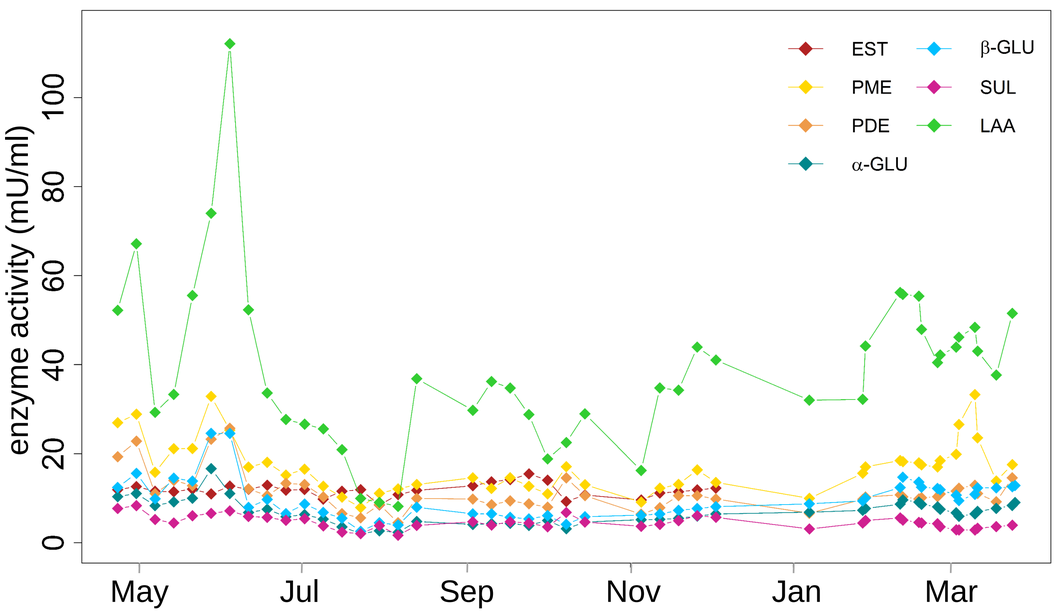
- All process stages except biological treatment: N_{tot} (mg/l), NH_4-N (mg/l), P_{tot} (mg/l), TOC (mg/l), BOD_5 (mg/l).
- Biological treatment: dry matter content (g/l), sludge volume (ml/l).
- WWTP effluent: NO_3-N (mg/l), NO_2-N (mg/l).

Tab. 1 Applied assays for the photometric determination of enzyme activities.

enzyme	EC number	dilution ratio[a]	substrate	incubation time [min]	incubation temperature [°C]	stop reagent[b]	wavelength [nm]
EST	3.1.X.X	1:10	Fluoresceine diacetate	60	21	/	490
PME	3.1.3.X	1:5	4-Nitrophenyl phosphate	15	30	Na_2CO_3	405
PDE	3.1.4.1	1:5	Bis(4-nitrophenyl) phosphate	15	30	Na_2CO_3	405
α-GLU	3.2.1.20	1:5	4-Nitrophenyl α-D-gluco-pyranoside	30	30	Na_2CO_3	405
β-GLU	3.2.1.21	1:5	4-Nitrophenyl β-D-gluco-pyranoside	30	30	Na_2CO_3	405
LAA	3.4.11.2	1:5	L-Alanine 4-nitroanilide hydrochloride	15	30	TCA	405
SUL	3.1.6.1	/	Potassium 4-nitrophenyl sulfate	15	30	Na_2CO_3	405

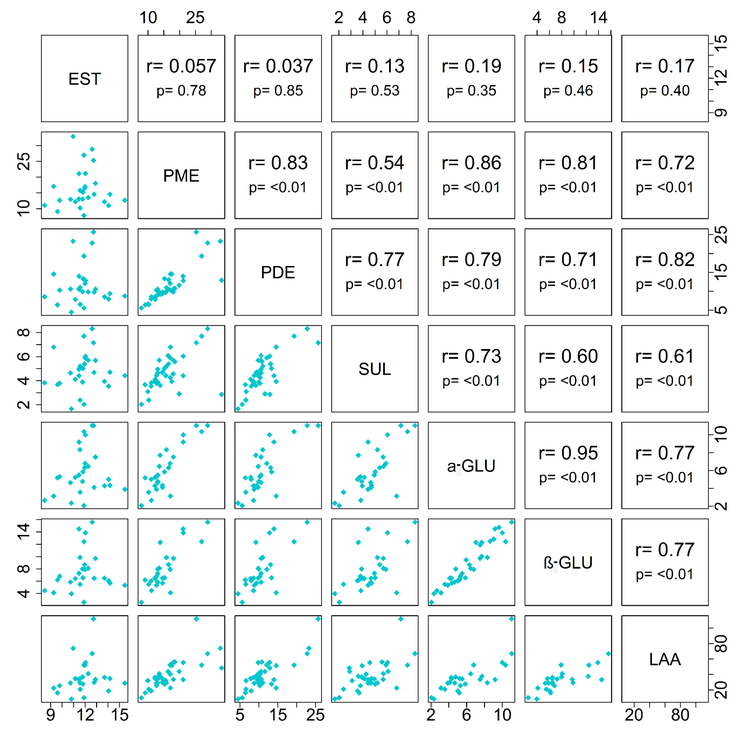
[a] Dilution in 0.14 M aqueous NaCl solution prior to assay. [b] reagents' concentration: Na_2CO_3: 1 M, TCA: 0.6 M

Results



Left: Fig. 2 Enzyme activities (mU/ml) in one subunit of the biological treatment stage of the WWTP Trier during the course of a year (April 2014 – March 2015).

Right: Fig. 3 Intercorrelations of enzyme activities (mU/ml).



Tab. 2 Spearman's coefficients for the correlation of enzyme activities with wastewater and process parameters. n = 47 - 48, exception: EST: n = 28, no asterisk p-value ≥ 0.05; * p-value < 0.05; ** p-value < 0.01.

enzyme activity			PME	PDE	SUL	LAA	α-GLU	β-GLU	EST
influent	**NH_4-N**	mg/l	0.26	0.21	0.35*	0.40*	0.26	0.22	0.53**
	N_{tot}	mg/l	0.17	0.15	0.40*	0.24	0.16	0.15	0.57**
	P_{tot}	mg/l	0.42*	0.47**	0.43*	0.49**	0.44*	0.42*	0.39
primary clarifier effluent	**P_{tot}**	mg/l	0.40*	0.42*	0.34*	0.32	0.47**	0.48**	0.46*
	TOC	mg/l	0.21	0.37*	0.30*	0.26	0.41*	0.38*	0.25
biological treatment	**DM**	g/l	0.73**	0.65**	0.39*	0.58**	0.60**	0.59**	0.45*
	SV	ml/l	0.51**	0.58**	0.58**	0.38*	0.44**	0.40*	0.26
effluent	**TOC**	mg/l	0.56**	0.59**	0.39	0.58**	0.63**	0.57**	0.46*
	TOC/TON	/	0.43*	0.43**	0.34	0.36*	0.47**	0.43*	0.16
input-output balance	**Δ N_{tot}**	mg/l	0.31	0.36*	0.32	0.40*	0.33	0.39*	0.48*
	Δ P_{tot}	mg/l	0.36*	0.42*	0.31	0.28*	0.38*	0.46**	0.45*

Tab. 3 Further correlations of EST activity with wastewater and process parameters. EST: n = 28, no asterisk p-value ≥ 0.05; * p-value < 0.05; ** p-value < 0.01.

influent	**BOD_5**	mg/l	0.41*
	TOC	mg/l	0.45*
primary clarifier effluent	**BOD_5**	mg/l	0.52**
	NH_4-N	mg/l	0.53**
	N_{tot}	mg/l	0.48*
effluent	**NH_4-N**	mg/l	-0.47*
	NO_3-N	mg/l	0.55**
input-output balance	**Δ NH_4-N**	mg/l	0.55**

Conclusions

- No influence of seasonal effects (e.g. temperature) on enzyme activities detected.
- LAA exhibited highest activity, SUL the lowest.
- Strong intercorrelations between certain enzyme activities due to similarity of enzyme-catalyzed reaction and substrate preference.
- Correlations of all enzyme activities with various process parameters and wastewater constituents, e.g. total concentration of P in various treatment stages, TOC content of effluent.
- Different activity pattern of EST resulting in lack of intercorrelations with activity of other enzymes.
- Deviating dependencies of EST activity on wastewater parameters.

} Concerns regarding the informative value of the EST activity as a sum parameter for hydrolytical enzyme activity.

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