

Using a translocation experiment with feral brown trout to investigate anthropogenic impacts on fish

Fabian G. Weichert^{1*}, Jörg Ahlheim², Werner Brack^{1,2}, Mario Brauns³, Markus Brinkmann⁴, Dorian D. Dörge⁵, Barbara Feldmeyer⁶, Patrick Fink^{3,7}, Markus Hecker⁴, Sarah Johann¹, Sven Klimpel^{5,6}, Martin Krauss², Markus Pfenninger⁶, Joachim Sturve⁸, Henner Hollert¹

*E-mail contact: weichert@bio.uni-frankfurt.de

¹Department Evolutionary Ecology & Environmental Toxicology, Faculty of Biological Sciences – Goethe University Frankfurt, Frankfurt am Main, Germany; ²Department of Effect Directed Analysis, Helmholtz Centre for Environmental Research – UFZ, Leipzig, Germany; ³Department of River Ecology, Helmholtz Centre for Environmental Research – UFZ, Magdeburg, Germany; ⁴School of the Environment & Sustainability and Toxicology Centre, University of Saskatchewan, Saskatoon, Canada; ⁵Integrative Parasitology and Zoophysiology, Faculty of Biological Sciences – Goethe University Frankfurt, Frankfurt am Main, Germany; ⁶Senckenberg Biodiversity and Climate Research Centre, Frankfurt am Main, Germany; ⁷Department of Aquatic Ecosystem Analysis and Management, Helmholtz Centre for Environmental Research – UFZ, Magdeburg, Germany; ⁸Department of Biological and Environmental Sciences, University of Gothenburg, Gothenburg, Sweden

Introduction

- Waterbodies are impacted by **contaminants in complex mixtures**
- Ecologically relevant effects** are difficult to assess
- Impact-oriented *in situ* studies using realistic exposure scenarios are scarce

Aims

- Investigating impacts of multiple chemical stressors on feral fish by
 - comparing **biomarker response patterns** of translocated and locally adapted populations
 - identify biomarkers responsive to **long-term & short-term** exposure
 - applying the **translocation design** as diagnostic tool

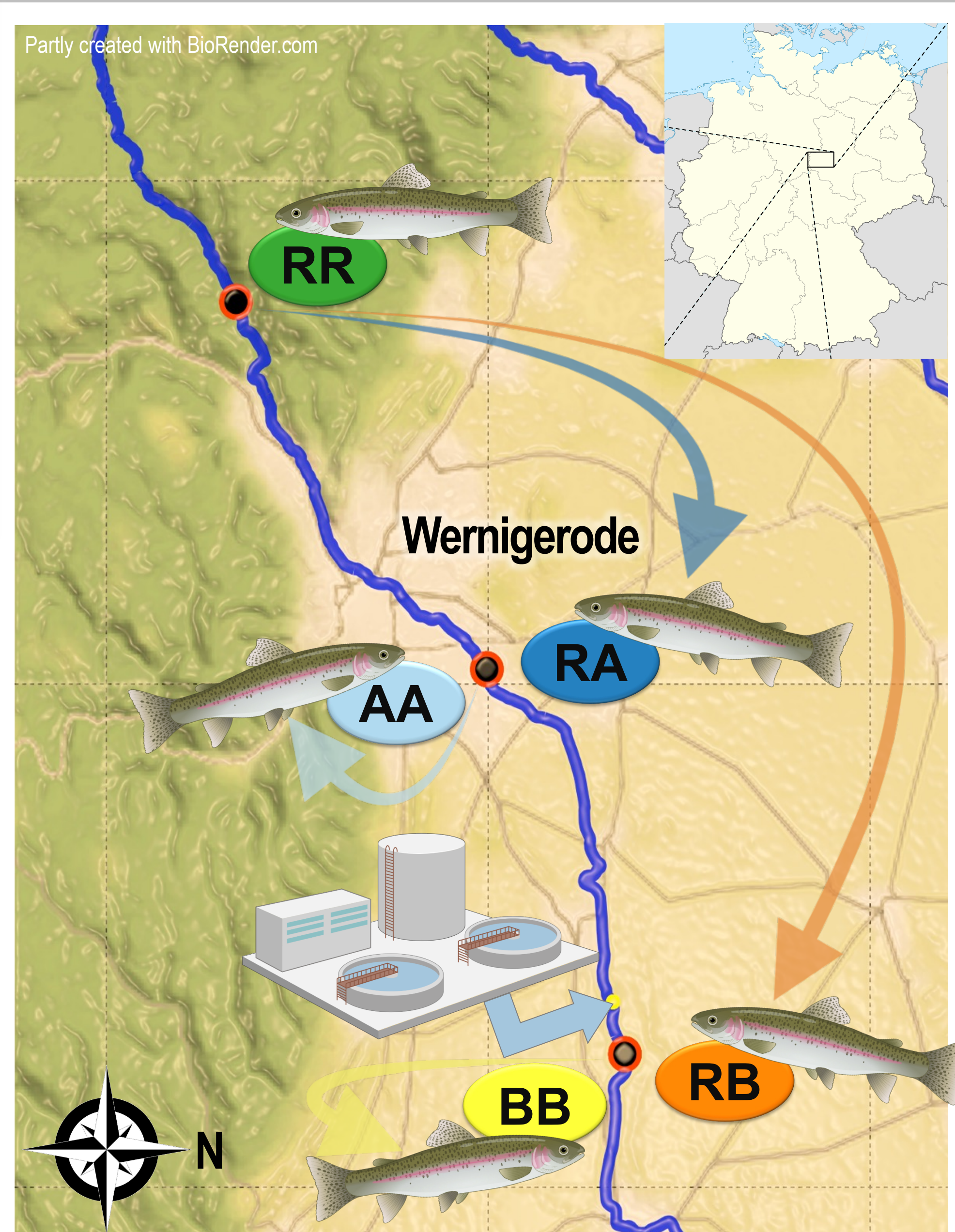


Fig 1. Schematic figure of the experimental design.

Experimental design & methods

- Brown trout (*Salmo trutta*) were caught at **three sites** and exposed (21 days) in **five groups**:
 - RR, AA & BB**: caught and caged at **reference site, urban site & downstream of WWTP**, respectively
 - RA & RB**: caught at reference site, **relocated to urban site & downstream of WWTP**, respectively
- Determination of haemoglobin (**Hb**), activity of **complement** system, acetylcholinesterase (**AChE**), liver somatic index (**LSI**), ethoxyresorufin-O-deethylase (**EROD**), glutathione-S-transferases (**GST**), glutathione reductase (**GR**), catalase (**CAT**), reduced (**GSH**) and oxidised glutathione (**GSSG**)
- Water sampling (large-volume solid phase extraction) & HR LC-MS/MS (457 compounds)

Results

	RR	AA	BB	RA	RB
Hb	-				
Complement	-	***			
AChE	-	*	**	*	**
LSI	-		**		
EROD	-	***		*	**
GST	-				**
GR	-		*		
CAT	-				
GSH	-			*	*
GSSG	-	**			*
GSSG/GSH	-	*	*		

Fig 2. Biomarker response patterns for locally adapted (AA & BB) and translocated (RA & RB) groups compared to the reference group (RR). Blue and red bars indicate increased and decreased response, respectively. The degree of change is shown by the length of the bar. Asterisks denote statistically significant differences ($p < 0.05$) using ANOVA + Holm-Šidák (Hb, AChE, GST, GR, GSH) or Kruskal-Wallis + Dunn's post hoc test (Complement, LSI, EROD, CAT, GSSG, GSSG/GSH).

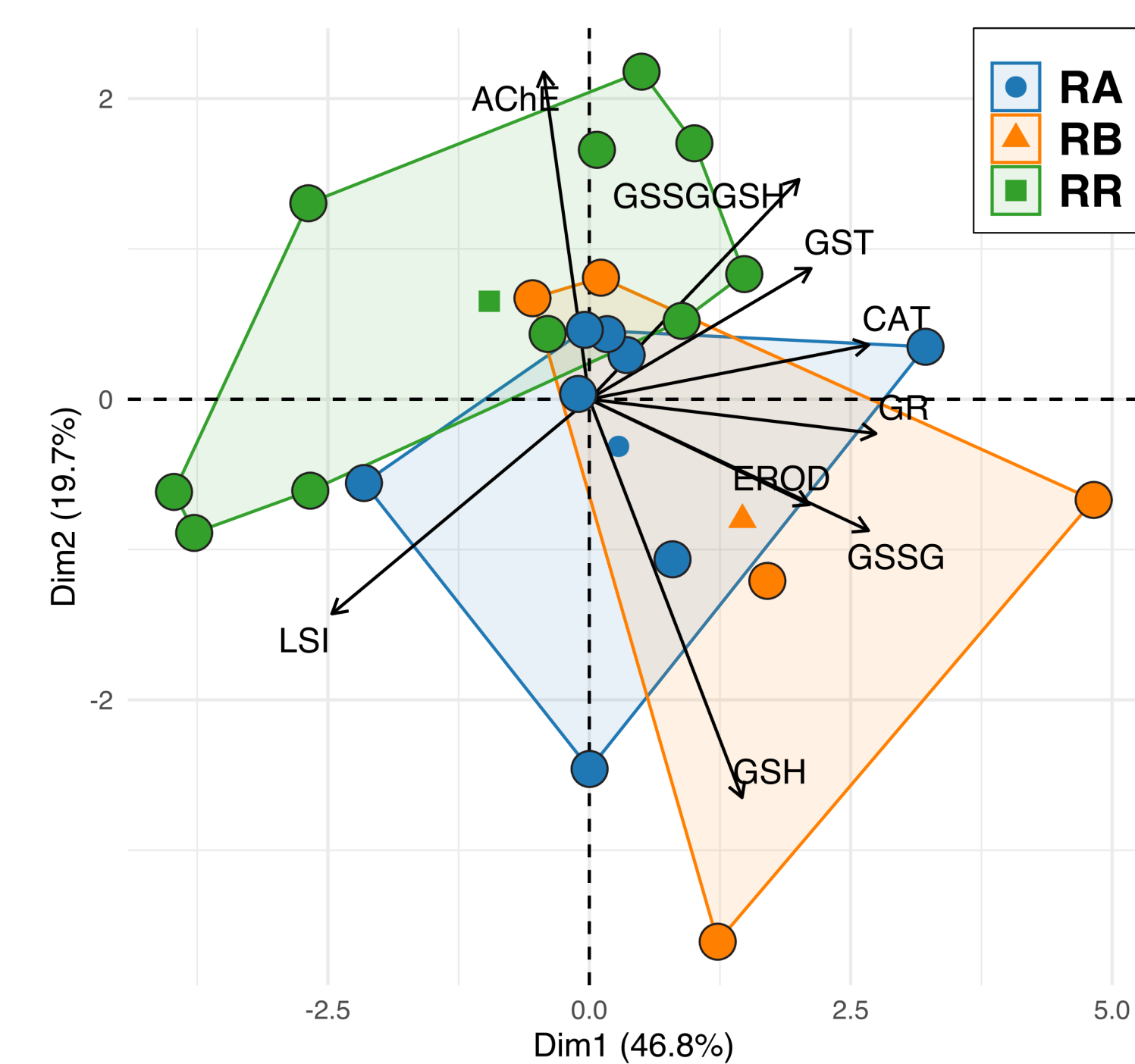
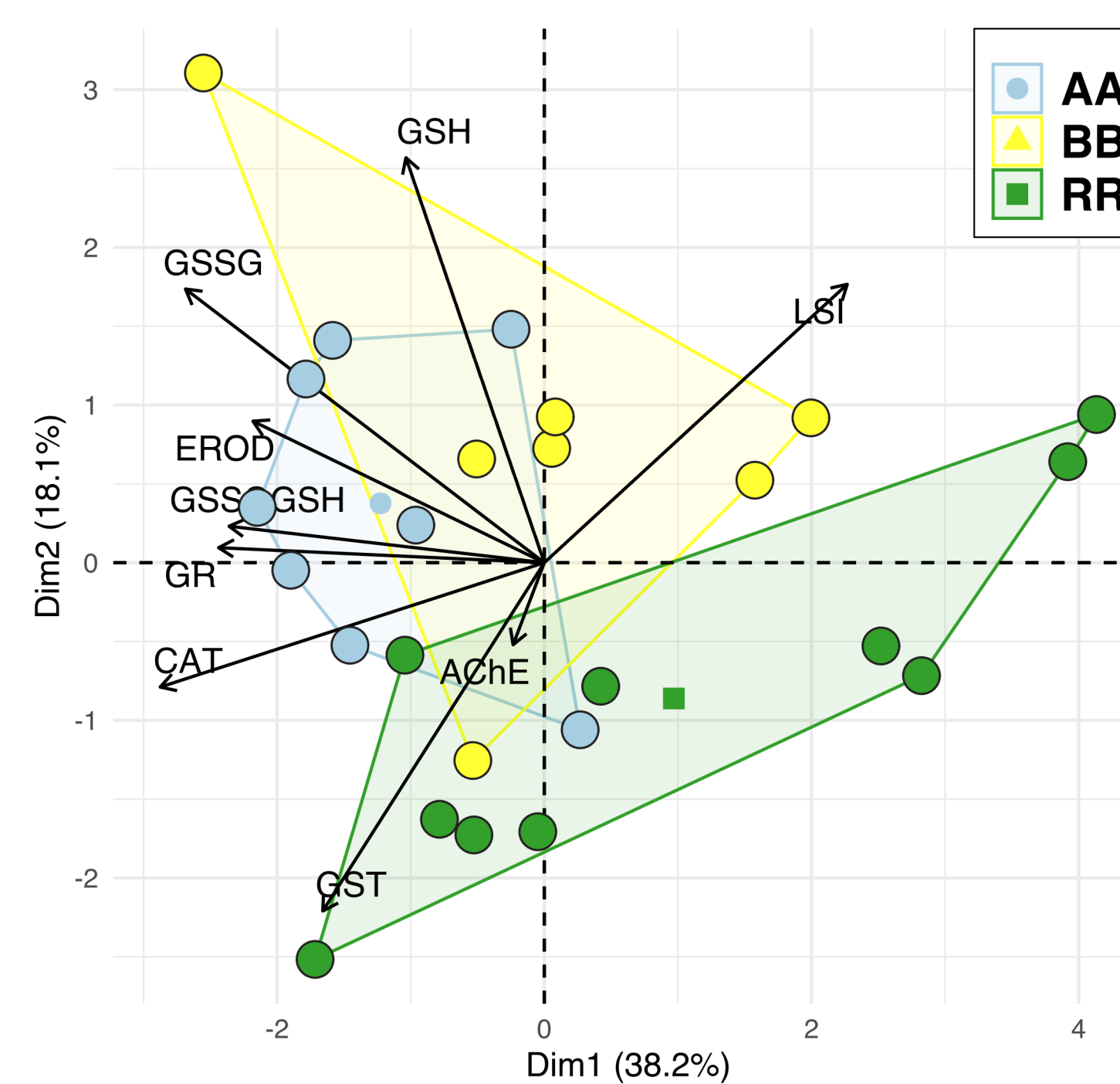


Fig 3. Principal Component Analysis of biomarker data (AChE, LSI, EROD, GST, GR, CAT, GSH, GSSG, GSSG/GSH) measured in female fish from reference & locally adapted groups (left panel) as well as from reference & translocated groups (right panel). The data set was reduced to female fish as GR, GSH and GSSG exhibited significant differences between male and female individuals.

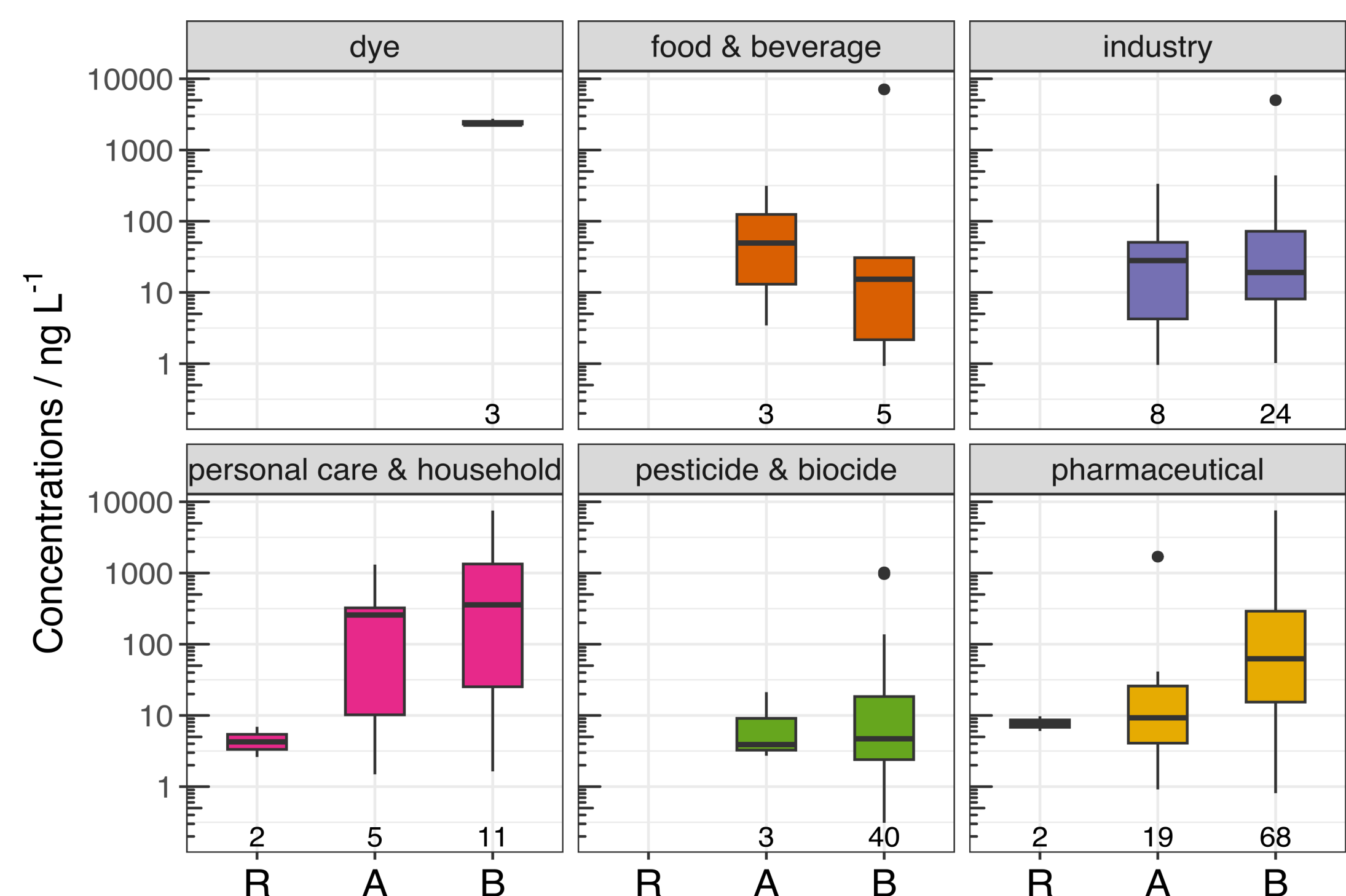


Fig 4. Measured concentrations of organic contaminants in extracts from each sampling site, grouped by compound class. Black horizontal dash shows the mean concentrations. Numbers above sampling sites indicate number of chemicals measured above method detection limit.

Conclusion & Outlook

- Biomarker responses vary between locally adapted and translocated populations
- Short-term** responsive: GST & GSH
- Long-term** responsive: LSI, GR, GSSG/GSH
- Overall** responsive: AChE, EROD, GSSG
- Next steps
 - RNAseq** data will be analysed
 - Molecular sex determination**
 - Determining **internal concentrations** of contaminants in fish tissue

Take-home message

- Pollution from **urban areas** and **WWTP** impact biomarker responses
- Measuring **multiple biomarkers** is crucial for the understanding of exposure impacts
- Translocation experiments** are a great tool to **study adaptational processes**



in memoriam Otfried Wüstemann