



# Large volume extraction of WWTP effluent for multiple bioassays

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# Method and Materials

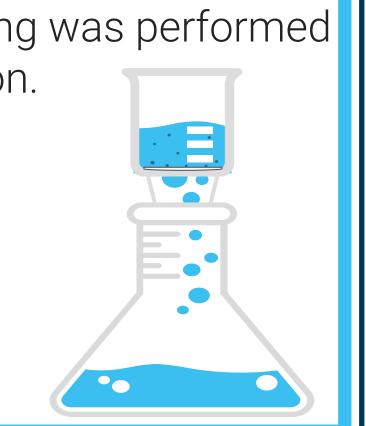
## Introduction

In the Netherlands, pharmaceutical analysis and a panel of CALUX® bioassays are used to asses the effectiveness of additional treatment steps in waste water treatment plants (WWTP).

Large number of assays ask for large extract volumes. The Water Laboratory investigated an automated SPE system to perform these LV extractions.

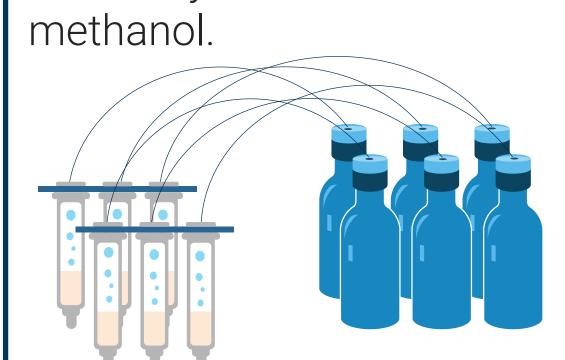
### Filtration

To rid WWTP effluent of debris, vacuum filtration was performed using cellulose filters. Spiking was performed after filtration.



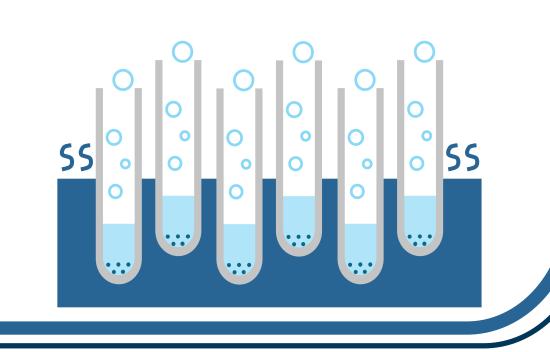
#### Extraction

Extraction and elution were performed completely automated. Elution solvents were ethylacetate and methanol.



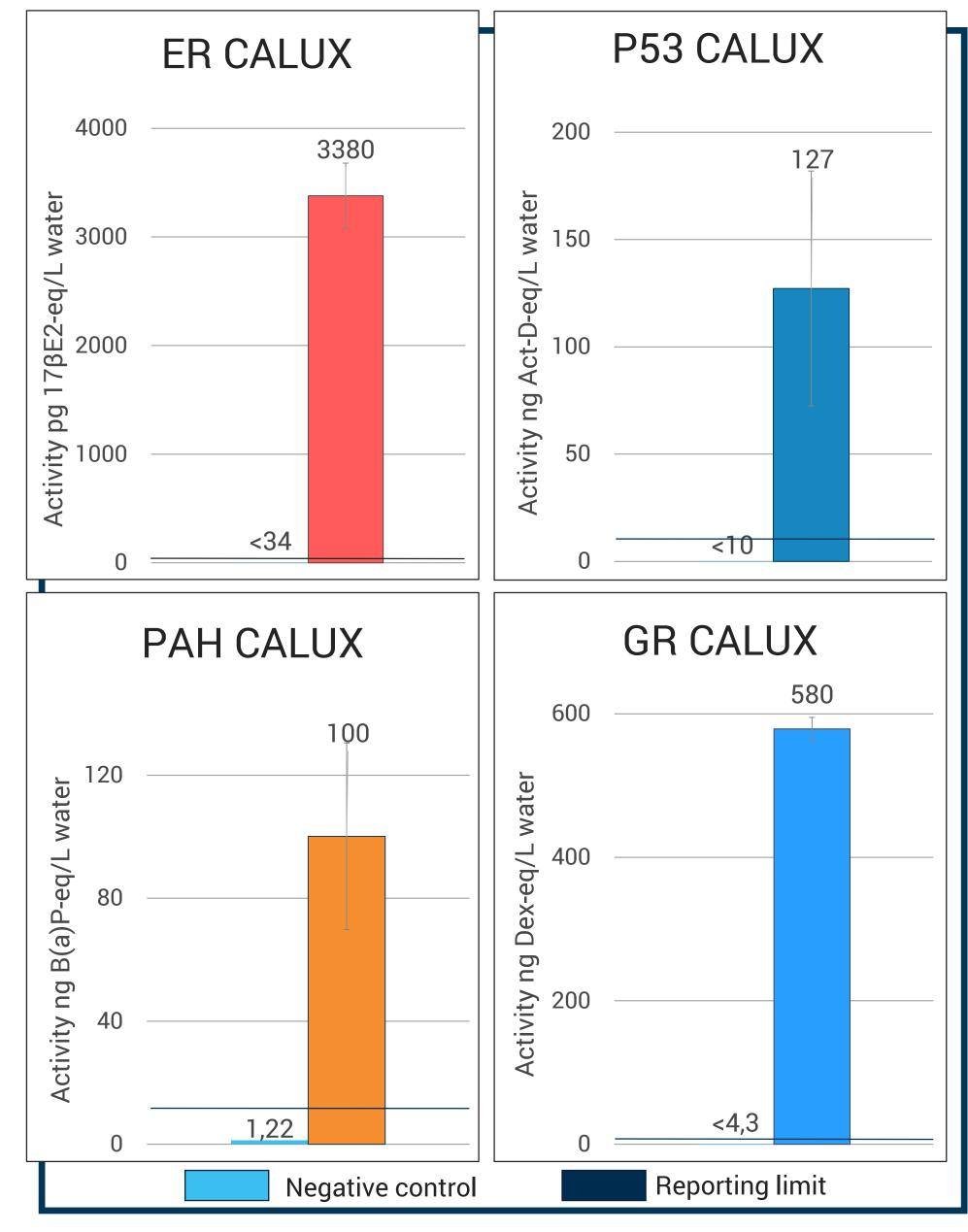
#### Concentration

The eluates were evaporated untill dry and then redissolved into DMSO (bioassays) or water (pharmaceutical analysis).



## Results

#### WWTP effluent and negative control

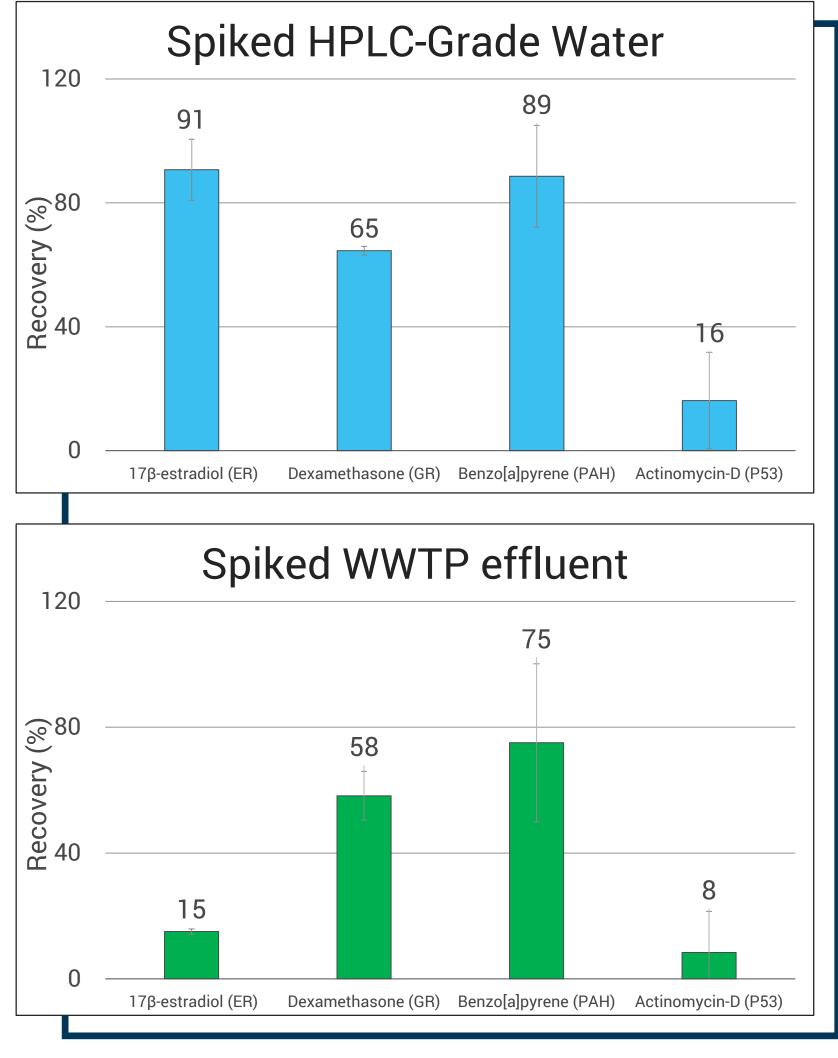


- Negative controls did not show activity in any of the assays.
- WWTP effluent showed activity far above the reporting limit in all assays.

# Acknowledgement

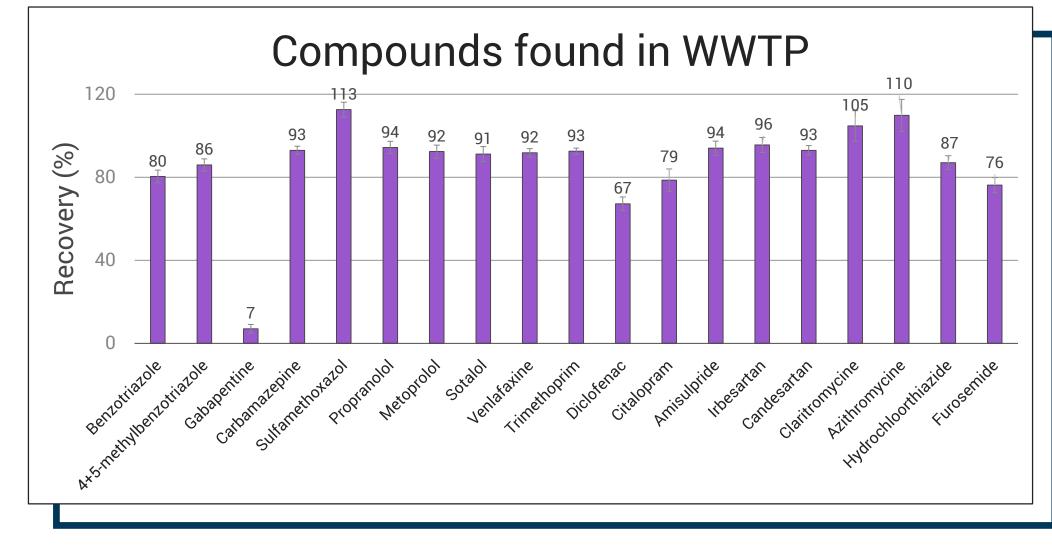
This work was performed in cooperation with the project "Advanced treatment or WWTP effluents" of the Department of Waterways and Public works of the Dutch Government<sup>1</sup>.

#### Spike recoveries



- Good recoveries (>60%) were achieved for ER, GR and PAH in HPLC-grade water.
- Lower recovery in WWTP effluent, probably caused by cartridge overloading.
- Actinomycin-D was not recovered efficiently.

#### Pharmaceutical analysis



 Pharmaceutical analysis showed high recoveries except for gabapentine. LV extraction method is thus capable of extracting a broad spectrum of compounds from WWTP effluent.

# Laboratory set-up

Gilson GX-274



## Conclusion

LV extraction on an automated system is a viable option for use in WWTP effluent and concentration of samples.

It is recommended to not go above 0,5 L effluent per SPE cartridge to prevent reduced recoveries in bioassays by matrix-overloaded cartridges.