

## INTRODUCTION

There is increasing recognition that there are limitations to the substance-specific approach for assessing and controlling the environmental fate and effects of effluents. Consequently, many regulators are seeking more holistic techniques such as whole effluent assessment (WEA) to supplement existing approaches. In general, WEA methodology assesses toxicity to aquatic organisms using whole effluent toxicity (WET) bioassays. The main objective of the project described in this, and two accompanying posters (Cailleaud et al. B, Glasgow 2013, Comber et al. C, Glasgow 2013), was to investigate the potential differences in outcomes for a risk assessment based on WET methodology, which is conservative, and one based on in-situ impact measurement. The difference between WET and in-situ impact measurement has been assessed using outdoor artificial stream mesocosms. This project has been designed and undertaken in three successive stages which were 1) experimental design and feasibility assessment; 2) understanding the biological responses in effluents and mesocosms and 3) comparing predicted, laboratory and mesocosm effects. The steps leading up to and including the final experiment are presented and discussed in this poster. The interpretation of the results will be presented in the two other posters.

### (1) How to transport effluents?

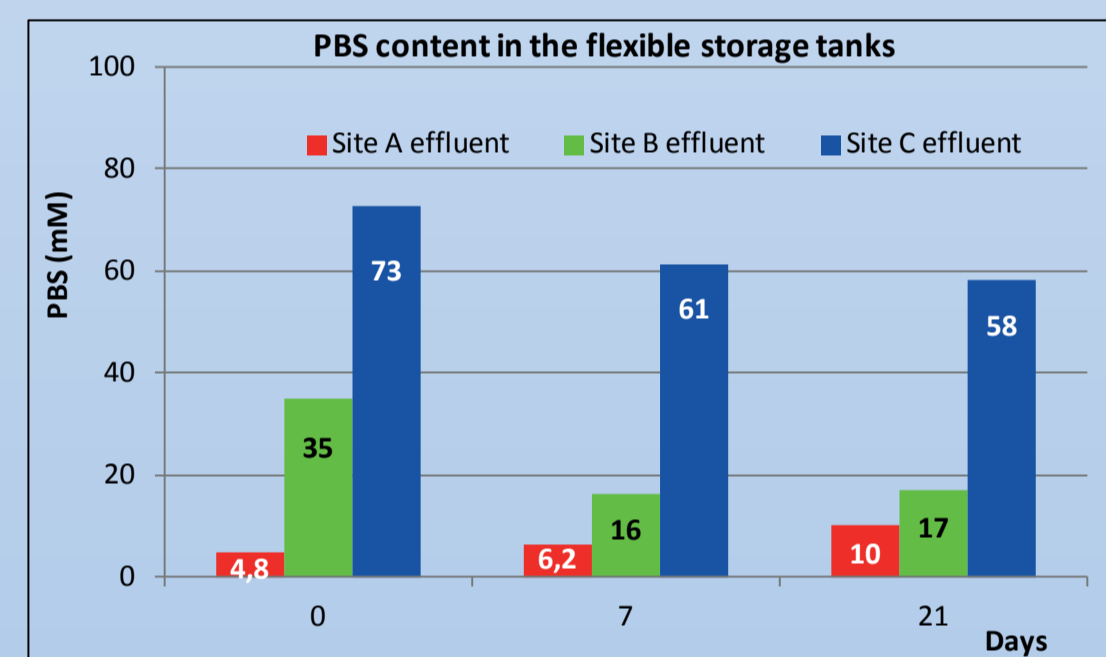
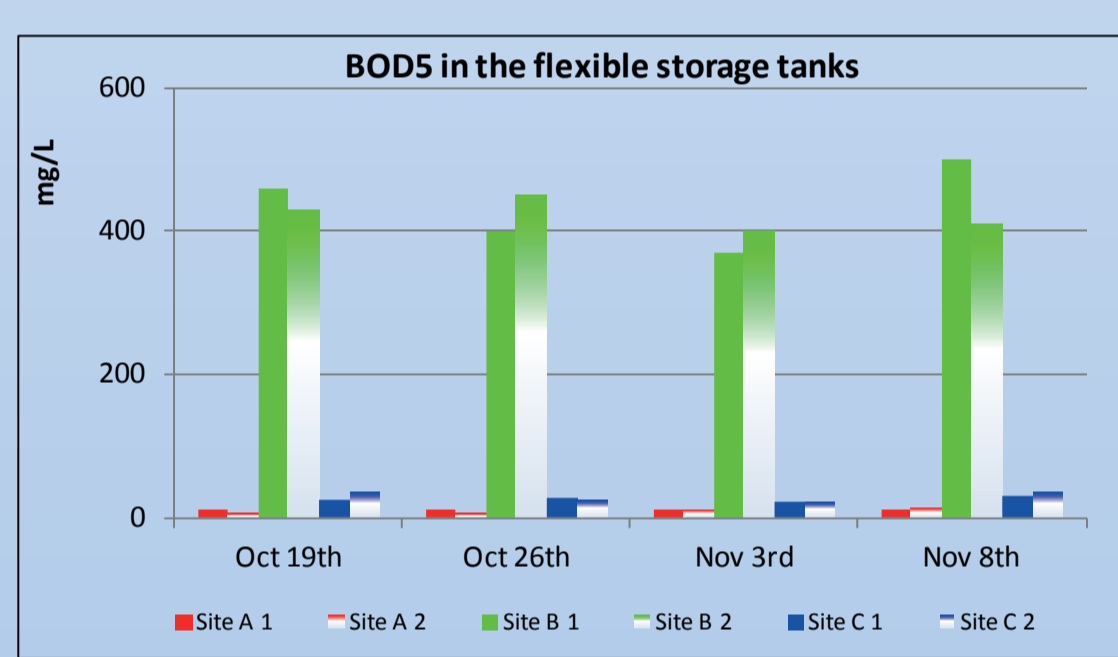
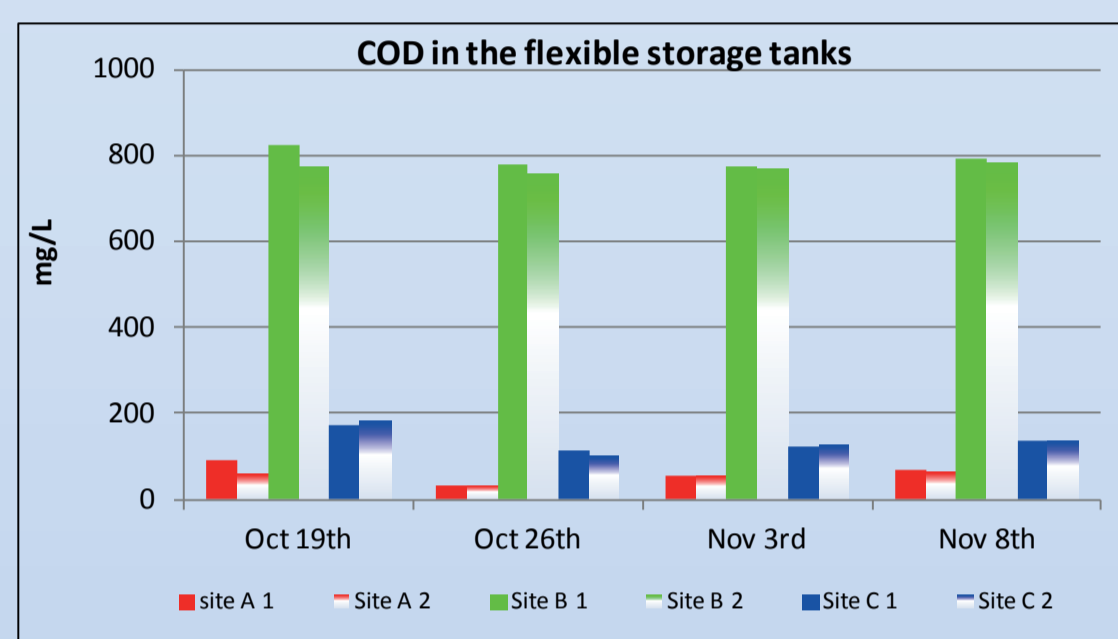
Transport: stainless trucks



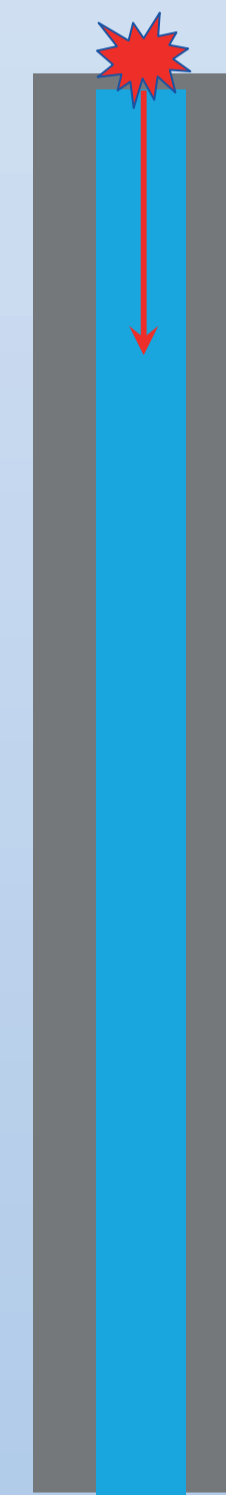
### (2) How to store effluents without aging?

Following extensive laboratory testing, it was determined that the best storage method was the use of flexible tanks made of plastomer-coated materials with no light or headspace. This ensured that the effluent was of consistent compositional quality.

Storage: flexible tanks

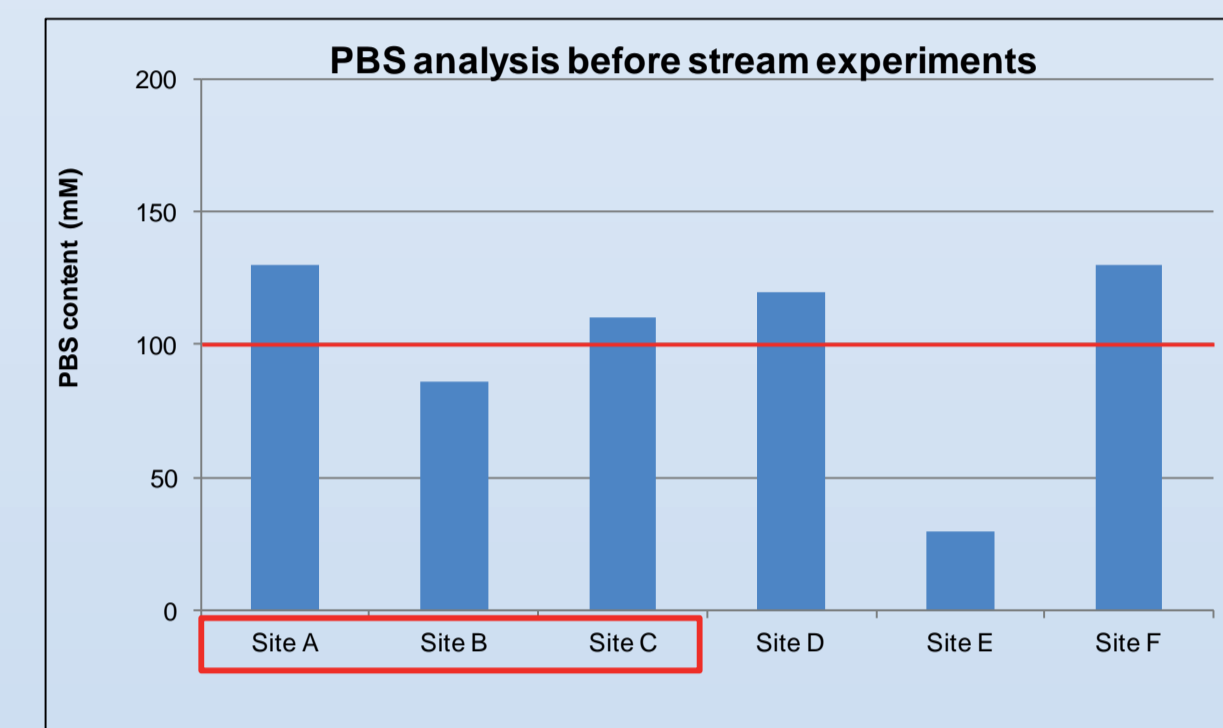


Injection system  
(Mixing valve)



### (3) Establishing the boundary conditions for observing effects

Indirect assessment of toxicity could be addressed by measuring the extractable hydrocarbons (PBS) obtained by solid phase micro-extraction (SPME), (Leslie et al., 2005). Based on literature review, a PBS threshold of 8 mM (Parkerton et al., 2001) should be present in the artificial streams in order to observe a chronic effect. Taking into account the minimum dilution factor in the artificial streams and PBS levels measured in various industrial sites, this threshold could not be maintained for a sufficient period of time with the available volumes of the effluents.



Endpoint	Critical C fiber (mM)	Reference
Acute narcotic effect trout	77	Parkerton et al., 2001
Acute narcotic effect algae	57	Parkerton et al., 2001
Acute narcotic effect <i>Daphnia magna</i>	42	Parkerton et al., 2001
Chronic narcotic effect <i>Daphnia magna</i>	8	Estimated based on ACR of 1/5*

Two options were therefore considered to reach 8mM in the streams for the duration of the experiment. The first was to decrease the flow rate of the streams and so reduce the required effluent volume. The second was to fortify effluents with an appropriate petroleum distillate so as to increase the PBS concentration and thus reduce the required effluent volume. A combination of these two options was used in the final experiments.

### (4) How to inject effluents + petroleum distillates into the streams ?



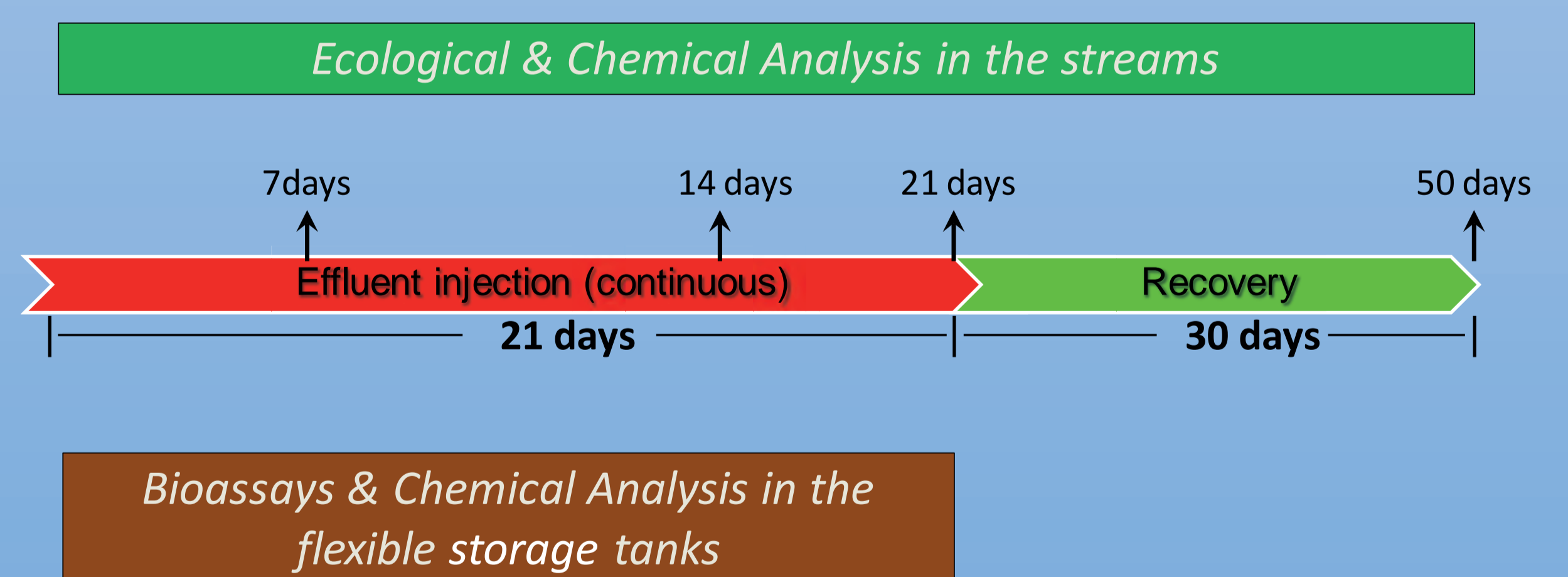
Specific equipment was used for the injection of the mixture of effluent and petroleum distillate. A mixing valve was used to improve dissolution of hydrocarbons in the water. In order to avoid volatilization of the distillate during the experiments, the headspace in the containers was filled with nitrogen.

## CONSEQUENCES FOR DEFINITIVE EXPERIMENTS

- Refinery effluents were sampled at three different sites (A, B and C) and tested in the streams.
- One effluent (A) was not fortified whereas the two others were fortified with petroleum distillates (respectively effluent B with Diesel and effluent C with Kerosene).
- A total of nine artificial streams were used (three control streams and two for each treatment)
- The streams were continuously treated for 21 days and during this time the parameters listed in the table below were measured once a week, both in the streams and in the flexible storage tanks.
- Thirty days after ceasing treatment, the recovery of the ecosystem was assessed on the basis of the same measured parameters.

The results of the definitive experiments are described in the second poster (Cailleaud et al. B, Glasgow 2013) and compared with those obtained using laboratory bioassays conducted on the whole effluents (Comber et al. C, Glasgow 2013). Conclusions are then drawn regarding the outcomes for risk assessments based on the two sets of results

	In situ in the streams	Effluents
<b>Bacteria</b>	Heterotrophic aerobic bacteria (MPN)	15/30 min Microtox acute toxicity (ISO 11348-3)
<b>Micro-algae</b>	Diatoms (biodiversity, abundance and IBD index)	72 h <i>Pseudokirchneriella subcapitata</i> chronic toxicity (ISO 8692)
<b>Invertebrates</b>	Benthic invertebrates (biodiversity, abundance and IBGN, EPT indices)	24 h <i>Daphnia magna</i> acute toxicity (ISO 6341)
<b>General physical and chemical measurements</b>		
	pH, O <sub>2</sub> , conductivity	pH, O <sub>2</sub> , conductivity, Chemical oxygen demand (COD), Biological oxygen demand (BOD5)
<b>Chemical analysis</b>		
	TPH	TPH
	PBS	PBS
	2D GC	2D GC



## CONCLUSION

- Preliminary laboratory studies identified the best condition for storing effluent containing hydrocarbons i.e. containers made of materials that limited adsorption of hydrocarbons and with no headspace or light. For the mesocosm experiments, flexible storage tanks made of plastomer-coated fabrics were selected to reproduce those storage conditions for the effluent on-site.

- An initial experiment was run in the streams to confirm the feasibility of sampling, transporting, storing and injecting effluents into the streams. An injection system incorporating a mixing valve was shown to be appropriate for obtaining homogeneous and reproducible exposure concentrations of the effluent hydrocarbons in the streams.
- Fortification of the effluent with a petroleum distillate fraction was shown to be necessary in order to achieve concentrations of hydrocarbons in the streams that were sufficiently high to induce observable effects on the biota.

## REFERENCES

- Cailleaud, K., et al., 2013b, Assessing petrochemical effluents using mesocosms - understanding the biological responses, SETAC Glasgow  
 Comber, M., et al., 2013c, Assessing petrochemical effluents using mesocosms: Comparison of predicted toxicity and laboratory and mesocosm measured toxicity of petrochemical effluents, SETAC Glasgow  
 Leslie, H., et al., 2005, Prioritization in WEA: Screening whole effluents using solid-phase microextraction, SETAC Lille  
 Parkerton, T., et al., 2001, Biomimetic extraction as a cost-effective analytical tool for determining the aquatic toxicity hazard of complex petroleum products, SETAC, Madrid