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Analysis of Nonylphenols in Contaminated Soil and Sediment in the Mölndal River

MARTIN VELEZ LOZANO

Chemical Environmental Science and Water Environment Transport
Chalmers University of Technology
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ABSTRACT

Sustainable development in society means a global compromise to decrease, control and eliminate all sources of pollution involved in the environment. Many solutions to solve the problems involved with the organic contamination of soils have been presented. In this thesis the nonylphenol contamination of soil in an industrial area close to the Mölndal river have been studied. The nonylphenols are transported to the aquatic environment, and cause problems in the ecosystems because they are toxic and have the ability to bioaccumulate. Consequently, it is important to understand the transport mechanisms between soil and aquatic media (rivers, lakes, wells) and sediment. It is also important to develop analytical methods that ensure a good reliability and repeatability.

The first part of the thesis comprehends a literature study of the origin, transport and final fate of nonylphenols in contaminated soil, together with a risk assessment. In the analytical part of the thesis a new technique (microwave digestion) to extract nonylphenols in soil is compared with the commonly used Soxhlet method. The nonylphenols are analysed by gas chromatography after solid phase microextraction of the aqueous extracts. Flame ionization or mass spectrometry was used for the detection. Nonylphenols in soil samples at the former Akzo Nobel plant in Mölndal and in water and sediment along the Mölndal river were analysed.

The results obtained show a good agreement between the microwave digestion method and the Soxhlet method. The chromatographic analyses also indicate the presence of many other pollutants than nonylphenols. The levels found in the soil and sediment samples were in the range 6.4 - 69 and 10 - 98 mg/kg dry weight, respectively. The highest concentration of nonylphenols in the sediment in the Mölndal river, 98 mg/kg dry sediment, was measured at the ICA Maxi site. High concentrations of nonylphenols, 17 mg/kg dry sediment, were also measured in the sediment at Mölndalsbro, a location upstream the former Akzo Nobel factory, and indicate an upstream transport of polluted sediment or another source of pollution. No presence of nonylphenols was detected in the water samples. The existing methods to remediate the contaminated soil suggest the use of biochemical methods.

ACKNOWLEDGEMENTS

First I would like to thank my supervisor, Ann-Margaret Strömvall for her support, trust, knowledge and work which made this project possible.

I also thank the assistance of Jesper Knutsson and Olle Ramnäs for their collaboration and co-workers in the techniques described in this work. I extend all my gratitude to Chalmers and to Sweden to offer me this possibility to achieve a master's degree.

I really appreciate all the support from my friends wherever they are, all my gratitude is with them. Also I want to send a special thanks to my friends Farhan and Feda to share many great moments in Göteborg. I especially would like to thank my father, my brother and Josefina for all the support given during my higher education in Sweden.

This thesis work is dedicated to the most special people in my life: Guillermina and Ruth.

" Is simplicity best or simple the easiest
The narrowest path is always the holiest
So walk on barefoot for me suffer some misery
If you want my love..."

Martin L. Gore

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CHAPTER 1: INTRODUCTION

1.1 Description of the problem

The Mölndal river is situated in Göteborg, Sweden and about 12% of its catchments area is covered by lakes, and includes the municipalities of Göteborg, Mölndal, Partille, Lerum, Landvetter, Härryda and Borås. The Mölndal river starts from the lake Stensjön, flows through suburban and urban areas of the cities of Mölndal and Göteborg, and is then connected to the Göta river. The upper Mölndal river flows through a forest area, but also includes areas with various industries and factories. The river water quality has been affected by numerous industries situated along the river, emitting pollutants and introducing wastewater into the river through many activities as chemical manufacturing, paper industry, etc. Even though some industries have left the area or closed down, sediments containing their products are still likely to be found. A total of 10 000 tons of nonylphenols per year were produced at the Akzo Nobel Surface Chemistry factory in Mölndal and then sent for export or to the main factory in Stenungsund. Even if the plant in Mölndal was closed down 3 years ago, there are still high concentrations of nonylphenols in the soil at the former factory area, resulting from past leaching and spills. In this thesis work, water, sediment and soil samples were taken in July and September 2001 in different locations at the Akzo Nobel Mölndal plant and up- and downstream the river, to measure the levels of nonylphenols in these areas and to study transport processes from the contaminated soil to sediment along the river.

Before any decision will be taken to solve the present problem with nonylphenols in the Mölndal river, pollutants have to be measured. Measurement is a critical step correlated to sustainable development, because it is a step in which traditional analytical methods may pollute the environment by using toxic chemicals for analysis. Environment friendly measurements techniques are therefore necessary to assure sustainability. The analytical procedure has several steps: field sampling, sample handling, sample preparation, separation and quantification, statistical evaluation, decision-making and final remedial action. In all of these steps it is important to obtain as correct results as possible. Chromatographic methods such as gas chromatography (GC) and high performance liquid chromatography (HPLC) are widely used for analysis of organic pollutants in the environment. Solid phase micro-extraction (SPME) is well suited for the sampling of organic compounds. Gas chromatography coupled with mass spectrometry (GC-MS) allows detection of organic compounds at trace levels.

1.2 Aims and objectives

The objectives of this study are:

- To make an analysis of the information available to show the possible mechanisms of transport of organic pollutants and make them applicable in the field study on nonylphenols.
- To develop a sustainable development – oriented methodology for the analysis of the nonylphenols found in the samples.
- To develop a Microwave digestion method based on water extraction and compare it with the solvent based Soxhlet extraction.
- To describe a mechanism of transport of the nonylphenols through all the media: water, soil, sediment, and compare the results with earlier results.
- To suggest a possible solution (physical, chemical or biological) for a remediation of the terrain, water and sediment that involves pollution with nonylphenols.
- To make public the results obtained from the analysis and their interpretation of the Akzo Nobel Mölndal plant to the regional authorities.

1.3 Methodology for the thesis work

First of all, a study into the literature involved with theories of pollution transport was done. Transport mechanisms about transboundary pollution were considered. This study involves how soil, sediment and water could be affected by the contamination of nonylphenols. A comprehension of the methodologies for sampling was studied to get the type of sampling methodologies and the number of samples. For most of the soil and sediment samples taken in field, Soxhlet extraction was carried out and kept for further reference.

All the samples (both extracted with Soxhlet and microwave digestion) were analyzed by gas chromatography. The Soxhlet extracted samples were injected to a gas chromatograph with flame ionization detector (GC-FID) with a 10 µl syringe. The samples extracted with MWD were injected to the same apparatus with the help of a SPME device because they were in water solutions. Some of the samples were also analyzed on a gas chromatograph with a mass spectrometer (GC-MS) as detector to get the mass spectrums with more detailed information about the compounds found.

Standard solutions (both in water and solvent) of a pure nonylphenol product were analyzed by GC-FID and GC-MS, as standards for identification and calibration curves. Eight experiments were also carried out to determine the partition coefficient between the soil and water phases.

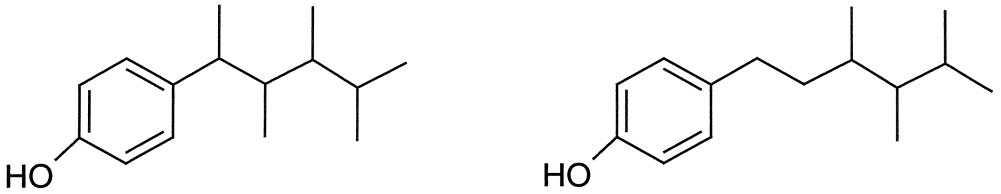
CHAPTER 2: BACKGROUND

2.1 The Akzo Nobel Mölndal plant

In this study, the source of nonylphenol pollution into the environment was the Akzo Nobel Mölndal plant with production of nonylphenol. The old factory was situated in Mölndal, Göteborg, Sweden and in operation during the period 1939-1999. At the factory nonylphenols were produced from alkylation of phenol with 1-nonene. The main reaction was according to the scheme:

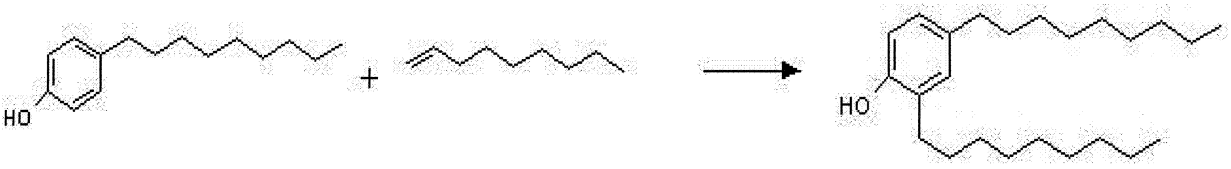
phenol 1-nonene *p*-nonylphenol

Because of the impurities of the raw materials, especially 1-nonene, which contains other isomers of nonene (branched isomers), by-products are obtained. The following are examples of other nonylphenols that could be obtained:



1,2,3,4-tetramethyl-*p*-pentylphenol 3,4,5-trimethyl-*p*-hexylphenol

Also di-alkylated product could be obtained when the product (p-nonylphenol) reacts with an excess of 1-nonene:



p-nonylphenol 1-nonene 2,4-dinonylphenol

Nonylphenols were sent to the Akzo Nobel plant at Stenungsund for production of nonylphenol ethoxylates, used as detergent, because of the surface-active properties (anionic and nonionic). Other uses of this compound are bactericides, lubricating oil, light stabilizers, agricultural chemicals, epoxy resin diluents, wetting and antifoam agents, dyes, drugs, adhesives, rubber chemicals, phenolic resins and plasticizer.

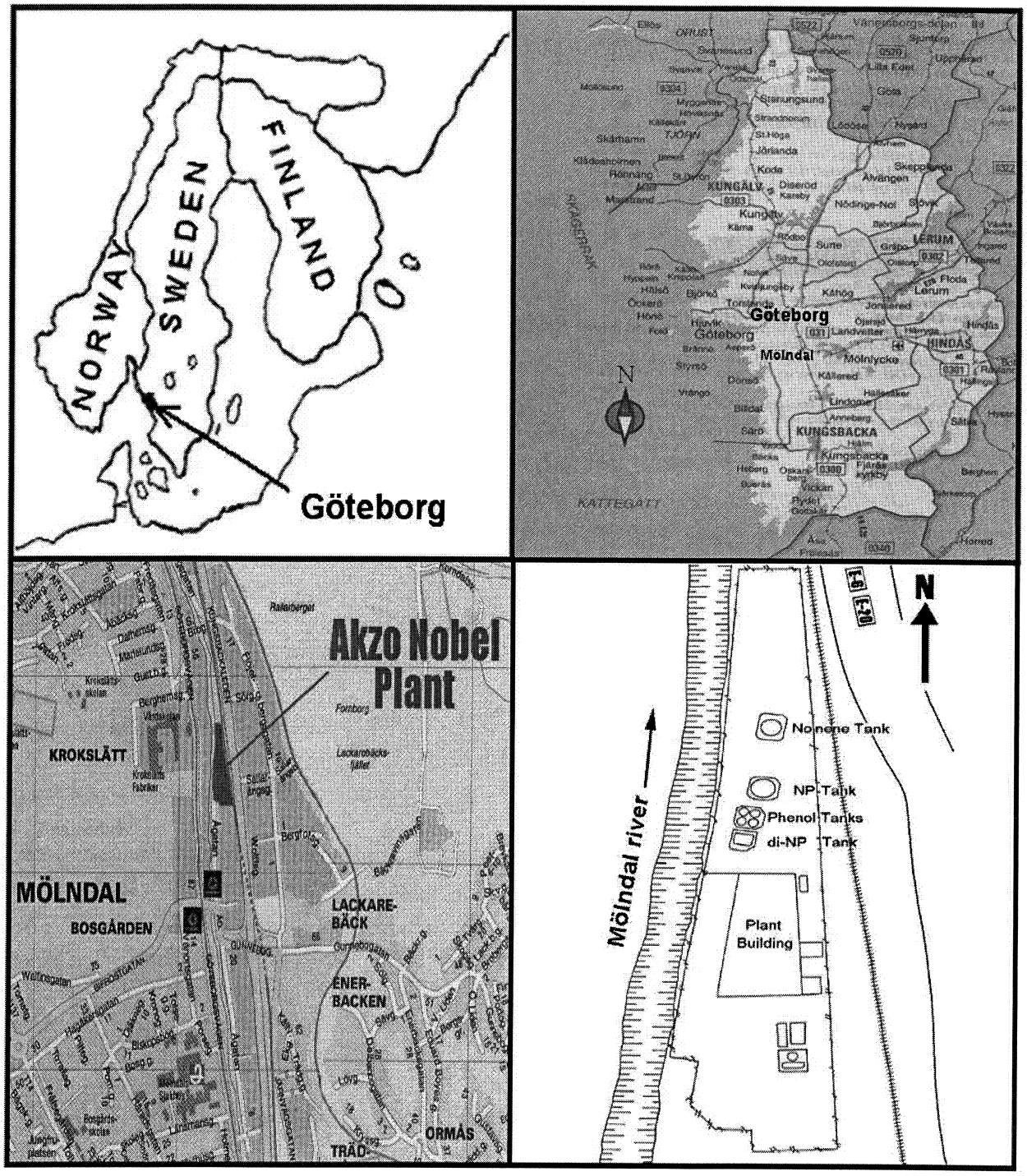


Figure 1: The location of the contaminated land at the former Akzo Nobel plant in Mölndal

CHAPTER 3:
MESUREMENT TECHNIQUES

3.1 The sampling procedure

The first step of the measurement of nonylphenols was to determine a suitable method for sampling. Samples were going to be taken in: water and sediment of the river and in soil at different locations at the Mölndal plant. For the sampling methodology, the methods chosen were recommended by IUPAC. In these methods there are several different strategies used to obtain a representative sample, presented in the Table 1.

Table 1: Different sampling strategies

Type of Sampling	Description
Probability sampling	
• Simple random sampling	Samples are selected using a random select system. All portions of a population have an equal chance of being selected.
• Stratified random sampling	Samples are selected from a carefully defined subpart (stratum) of the parent population. Within each stratum, the samples are selected randomly.
• Systematic sampling	Samples are chosen on a systematic basis, i.e. samples may be selected from a defined sampling area according to a grid design.
Non-Probability sampling	
• Judgmental sampling	The sampler uses personal judgment, experience and a professional criteria in selected samples from a population and area.
• Convenience sampling	Samples are selected on the basis of accessibility, expediency, cost or other personal reasons not directly concerned with statistical parameters.
• Restrictive sampling	Samples are taken from a part of population from a criteria that they are readily accessible to be quickly and easier chosen.

In this thesis work the non-probability sampling with the derivation of the judgmental criteria was chosen, according to a previous experience. In the field it is essential to select the appropriate sampling location and time. Further care must be exercised during sample collection to ensure that the sample integrity is not jeopardized. The sampling devices and procedures therefore have to be designed to avoid contamination, cross-contamination or losses due to adsorption (Haglund *et al.*, 1999). Glass, stainless steel and Teflon are widely recognized as the safest sample containers materials for organic pollution analysis.

The homogeneity of natural waters is similar to the ambient air, so the technique to collect the water samples is a simple immersion of a plastic (acrylic) Ruttner vessel. Samples of 0.5 - 1 liters were taken in this study. For the sediment sampling, a core sampler and a grab sampler were used to collect the samples in the bottom of the river at different locations. The grab sampler is a stainless steel box shaped recipient with two claws which are open until the box reach the bottom, then at willing, these claws can be closed and the sample picked up from the river. The approximate volume of the box was 5 liters, and the samples obtained were from 0.5-1 kg.

The uncertainties associated with the representativeness of the samples frequently exceed those inherent in the collection and analysis. Because of these problems it is best to collect as large amounts of sample as practically possible. Also a posterior cleaning of the samples is often recommendable, trying to remove all large stones, leaves, branches and possible garbage found in them. The procedure and equipment used in soil sampling is more simple. Often samples are merely dug out from a specific depth (30 - 40 cm in this work) and then transferred to a sample container. In case where the vertical distribution of contaminants is important, samples are taken using a core sampler (a metallic tube), that is punched into the ground. In this work a special digger (see Figure 2) used in geological studies were chosen, with a capacity of 200 - 300g in each dig, and samples were taken of 1 - 1.5kg each one.

After the samples were taken, they were put in the freezer immediately and were kept for a posterior analysis and extraction. The sediment and soil samples were saved into plastic bags and the water samples in plastic bottles with stoppers just to make freezing possible, but for short storage it is best to chose glass bottles because the plastics could adsorb nonylphenols. Before the extraction step, each sample was warmed in glass beaker until ambient temperature and then homogenized. Stones, branches, leaves, dead organisms and garbage were removed and soil and sediment samples were put into a porcelain mortar and homogenized by grinding. The samples were put into glass vessels and kept under normal refrigeration until they were extracted and analyzed. The water samples were carefully agitated after they melt.

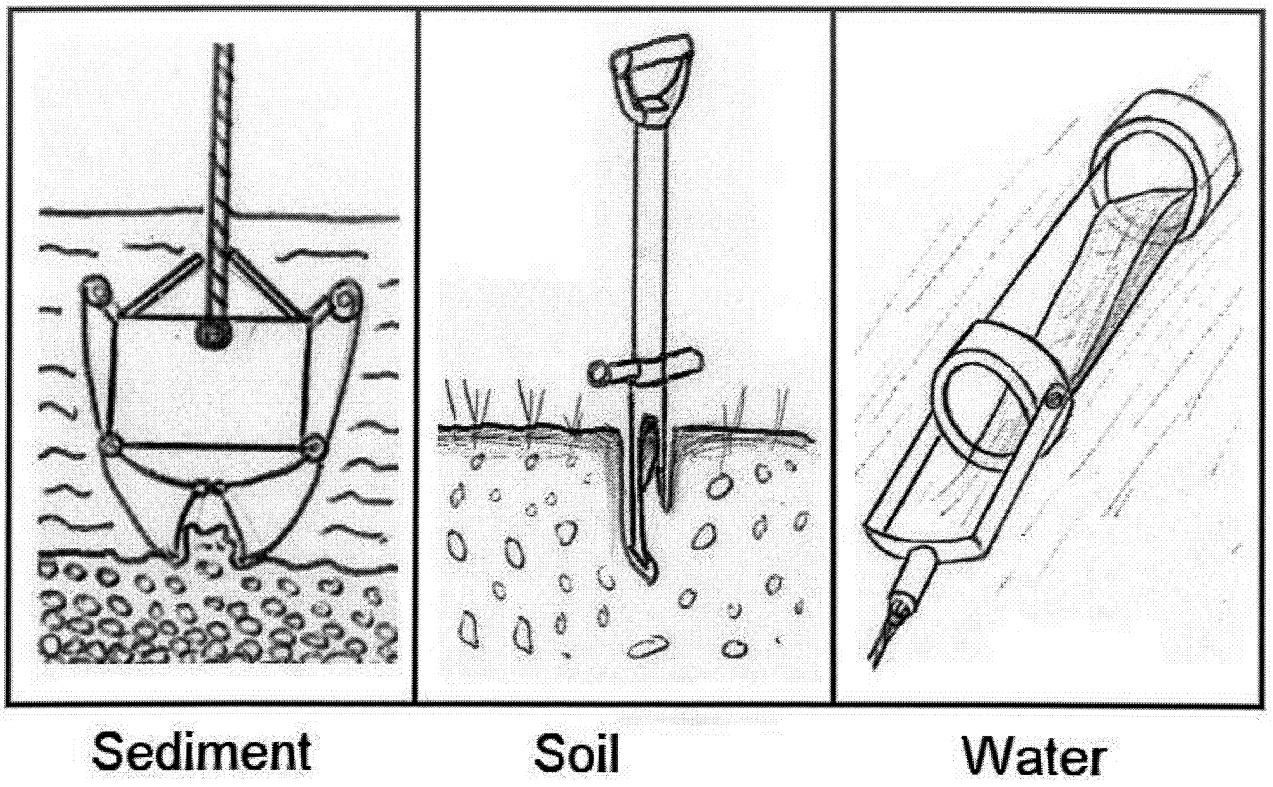


Figure 2: Samplers for soil, sediment and water

The plan for the location of the samples was as following:

- Sediment samples: 10 taken in September 2001 at the middle of the river (by boat) nearby (north, center, south) the factory plant. (No 1-10)
  2 taken in September 2001 down and upwards the river. (No 11,12)
  4 taken in July 2001 at the middle of the river (by boat) nearby (north, center, south) the factory plant. (No 13-16)
  3 taken July 2001 down and upwards the river. (No 17-19)
- Soil samples: 10 taken in September 2001 at 1-2 m from the shore nearby (north, center, south) the factory plant. (No 1-10)
- Water samples: 1 taken in September 2001 in the river from 5 km north of the factory plant. (No 1)
  3 taken in July 2001 in the river from 0.5-2.5 km north of the factory plant. (No 5,6,8)
  2 taken in September 2001 in the river from 0.5-5 km north of the factory plant. (No 5-9)
  3 taken in July 2001 in the river at the factory plant (No 2-4)

For the locations of the sampling points see Figures 3 - 6.

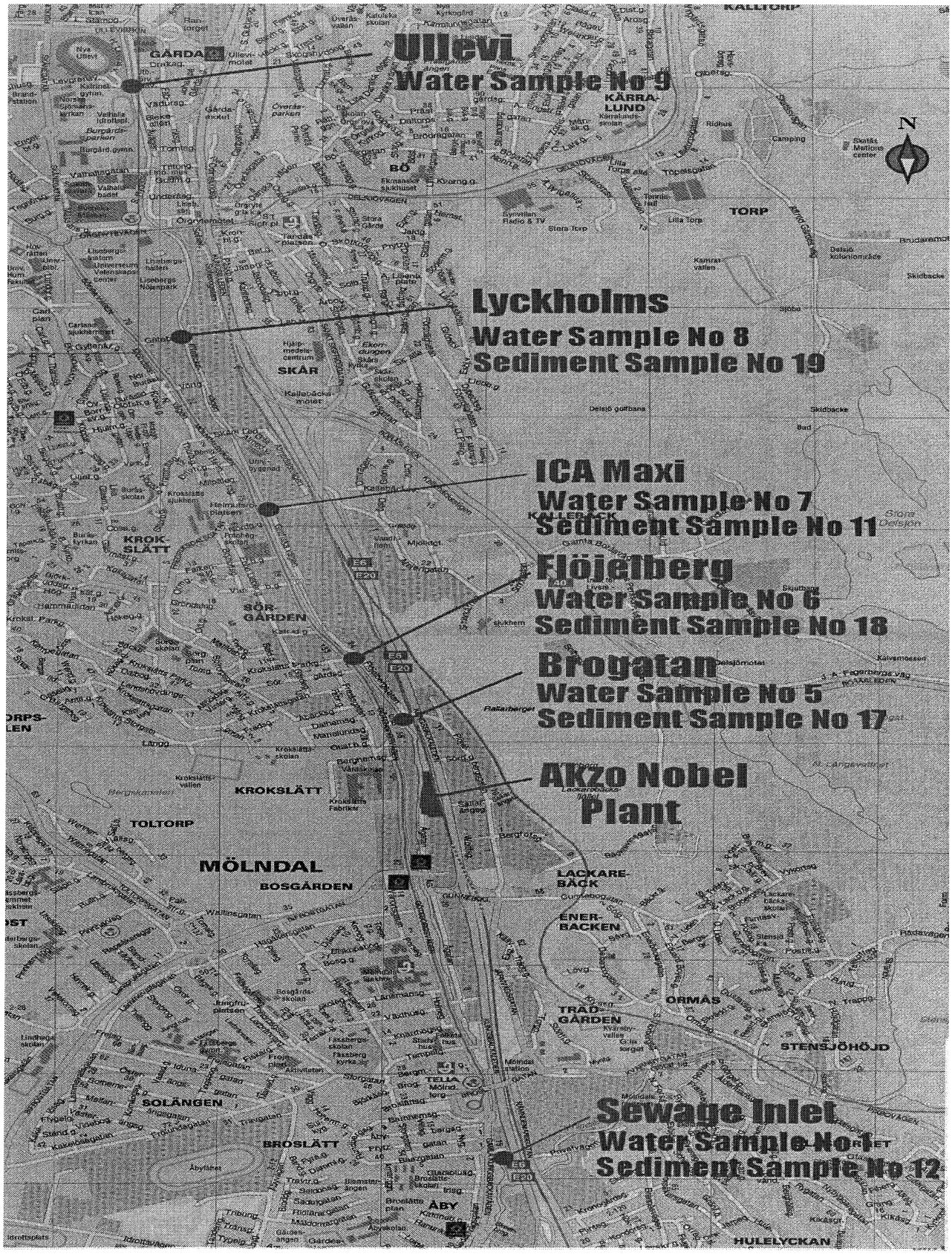


Figure 3: Location of the sediment and water samples taken in the urban area of the Mölndal river

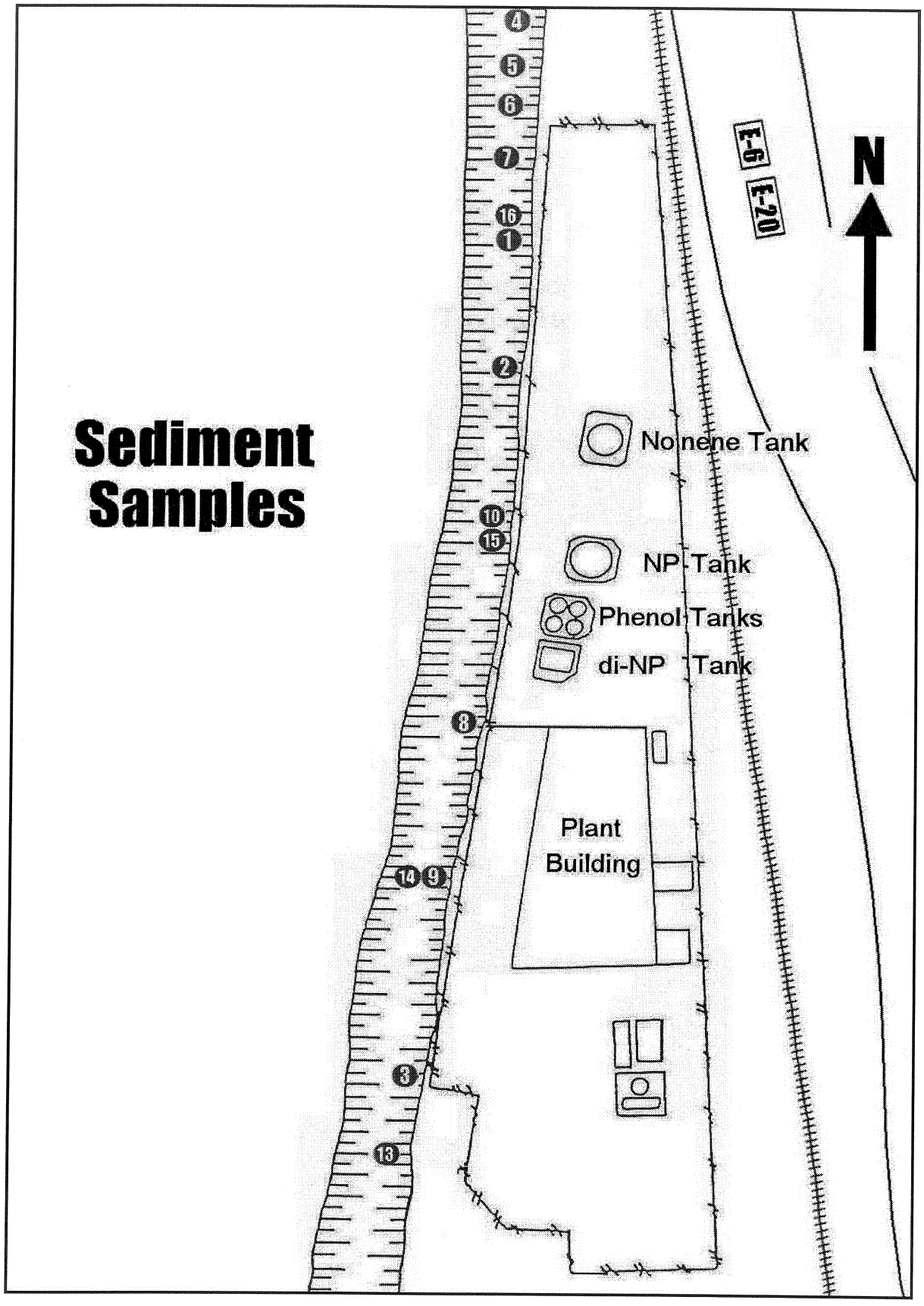


Figure 4: Location of the sediment samples taken at the Akzo Nobel Mölndal plant

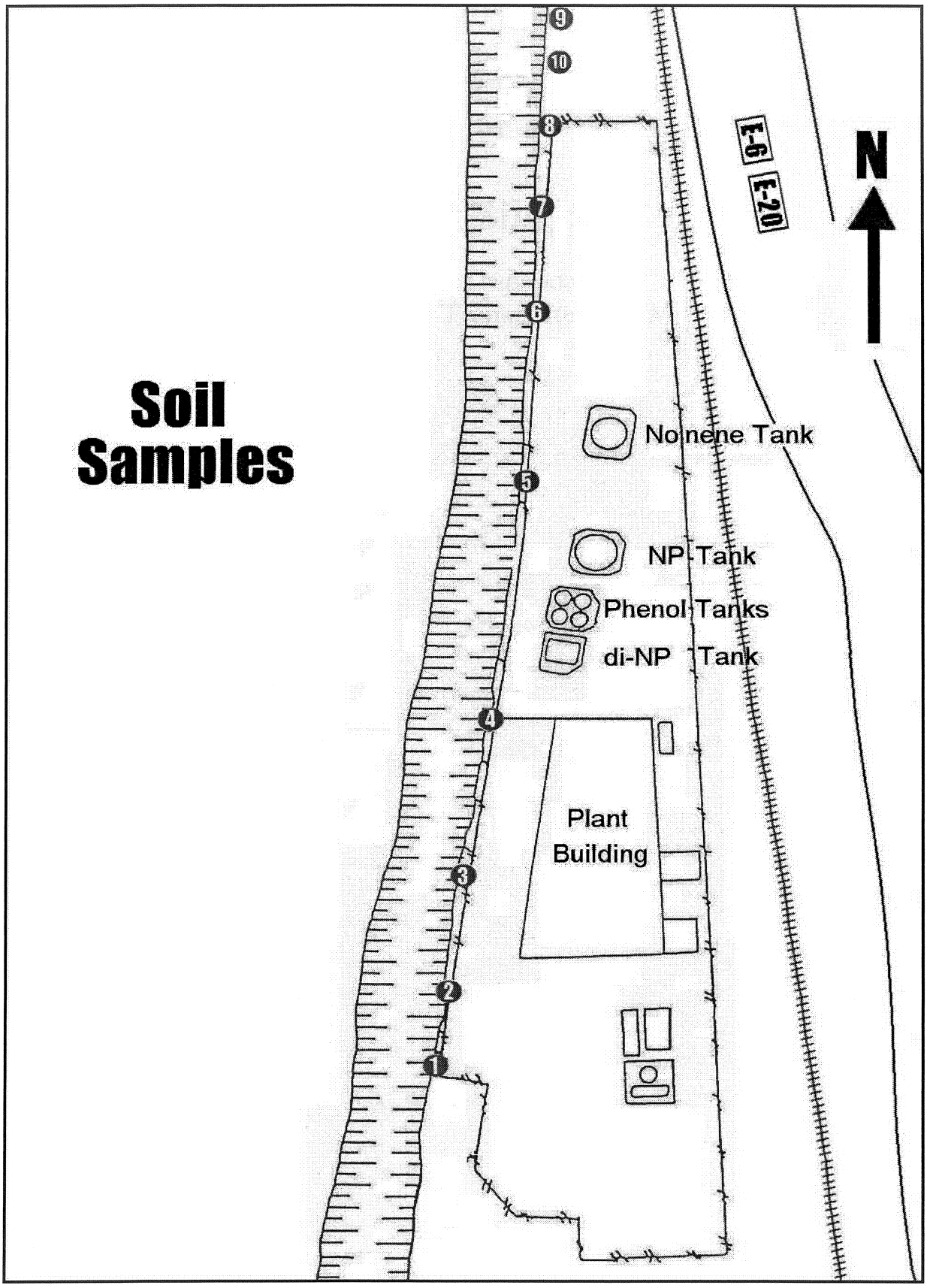


Figure 5: Location of the soil samples taken at the Akzo Nobel Mölndal plant

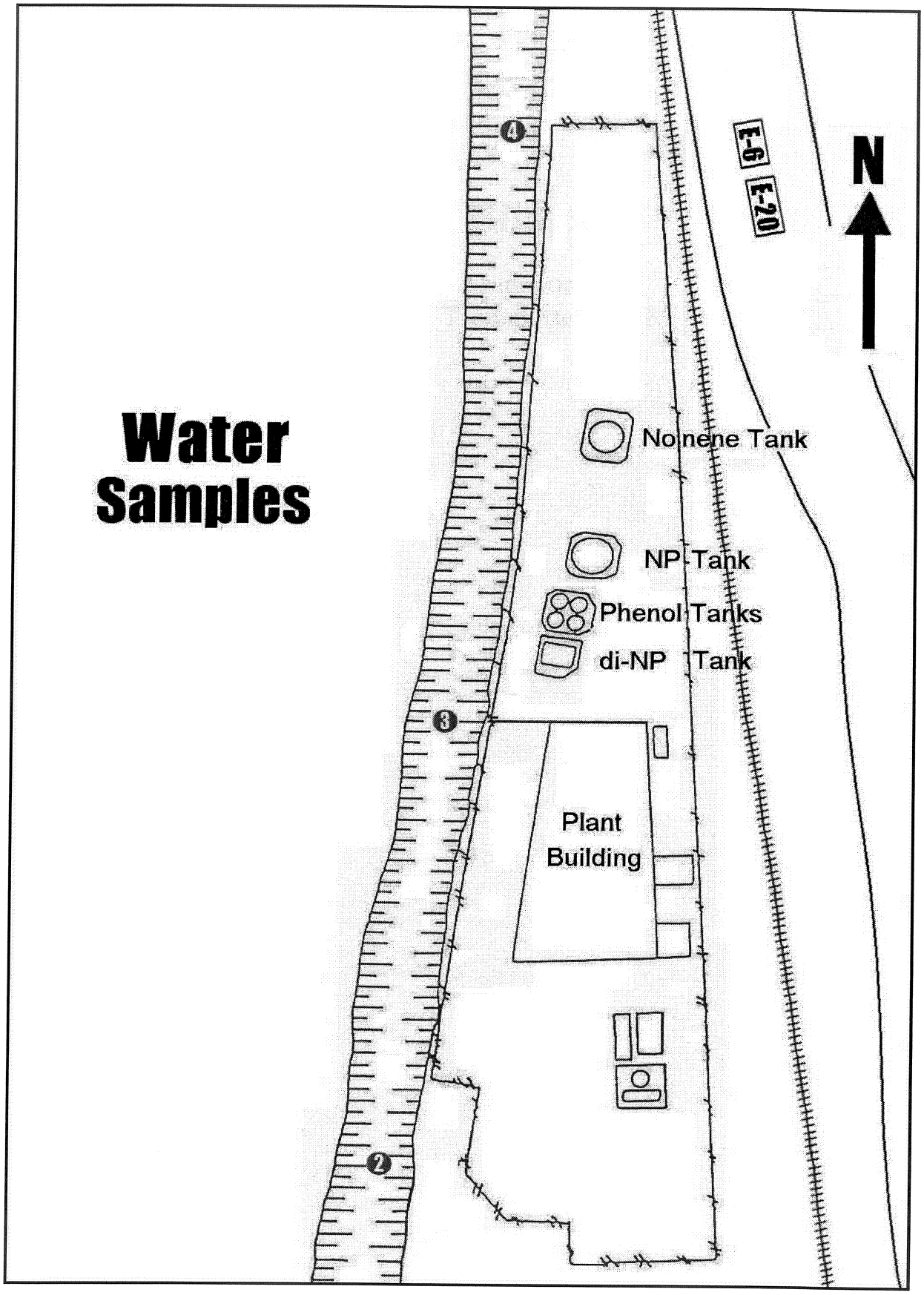


Figure 6: Location of the water samples taken at the Akzo Nobel Mölndal plant

3.2 Soxhlet extraction

This technique is based on the principle of the equilibrium between two phases (liquid-liquid or liquid-solid normally) and the solubility preference of one compound between two solvents or medias. The description of the equipment used for Soxhlet extraction is shown in Figure 7. The system consists of a vessel which contains a solvent to be boiled during the extraction. The solvent vapors are condensed in a cooler and drops fall into a column which contains the sample (soil or sediment) in a cellulose thimble. The condensed solvent remains drowning the sample and stays there for 3-5 minutes making an equilibrium between the solid phase and the solvent. At the end of this period, the solvent will be released by a siphon system into the Erlenmeyer flask which contains the boiling solvent. The solvent starts to evaporate again and the process is repeated. This process is repeated fully every 10 minutes and the whole process will take 24 hours. For this work, samples of ~20 g were extracted for a period of time of 24 hours (soil or sediment). At the end of the extraction process the solvent is collected. This solvent now contains all organic compounds earlier present in the solid (depending on the efficiency of the solvent in the extraction) and should be kept in a cool place for a posterior re-concentration. In this work *n*-heptane (450 ml) was chosen as solvent because it is one of the less toxic organic solvents for laboratory use. The equipment was cleaned between the extractions with n-heptane and acetone.

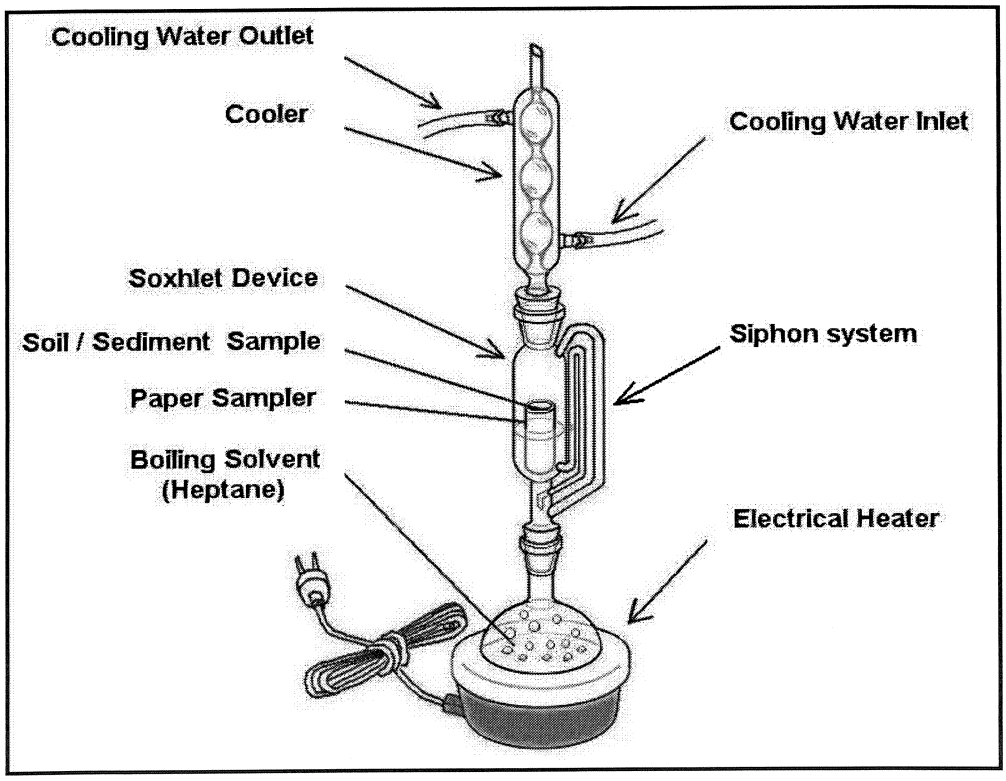


Figure 7: Description of the Soxhlet equipment

In the Soxhlet extraction, a solid sample is extracted with a solvent and the extraction process is a simple partitioning process. The extraction efficiency is dictated by the partition coefficients between the two phases:

$$K = \frac{[X]org}{[X]aq}$$

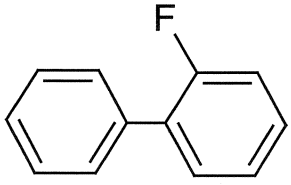
where K is the partition coefficient and $[X]_{org}$ and $[X]_{aq}$ are the equilibrium concentrations of a target analyte in the organic and aqueous phases, respectively. Solid samples may also be extracted with organic solvents. In this case the same type of partitioning process occurs, although it is a solid-liquid extraction and a similar relationship applies:

$$K = \frac{[X]s}{[X]org}$$

where $[X]_s$ is the equilibrium concentration in the solid phase.

Particulate matter (soil, sediment) often consists of an inorganic core covered by an organic surface layer, or simply the particles themselves contain a conglomerate of finer particles (Haglund *et al.*, 1999). The analytes might be sorbed both to the core and the organic surface layer, or might even be totally enclosed in the core. The analytes must undergo several processes before being extracted. These includes: A) a mass transport through an insoluble organic matrix to the matrix-fluid interface, and B) a partitioning at the matrix-fluid interface and bulk mass transport to the extraction medium. If the first process is the rate determining step, the extraction is kinetically controlled, but if the latter process is the rate determining step, the extraction is controlled by the particle surface-extraction medium partitioning coefficient. If the bulk of the organic solvent media is soluble the second process will dominate. In both cases, the extraction can be enhanced by grinding the sample, thereby, an increase of the exposed surface are and then decreasing the diffusion path length.

The addition of an Internal Standard (IS) is frequently used in the samples to monitor the extraction process and is used to compensate for losses of analytes during clean up, and to cancel the instrument instability. These standards may be added before, during or after the extraction process. In this project 1-fluoro-biphenyl was used as internal standard for the later gas chromatographic analysis (see Table 2):



1-fluoro-biphenyl

This substance was chosen because its mass spectrum is easy to interpret and one ion predominates.

Table 2: Amount of internal standard added to the samples

Sample Kind	Soil	Sediment	Water
Milliliters of a solution with a concentration of	0.5 ml 100 µg / ml	0.5 ml 100 µg / ml	0.5 ml 100 µg / ml
Total amount added (µg)	50	50	50

After the Soxhlet extraction and the collection of the concentrate solvent, the extracts were carefully evaporated. This step was carried out in a steam rotatory evaporator (Figure 8). In this equipment, the extract (≈ 400 ml) was put to boil in a hot water bath in a vacuum sealed system and then evaporated until a volume of 10 ml (volume obtained within 40-50 minutes).

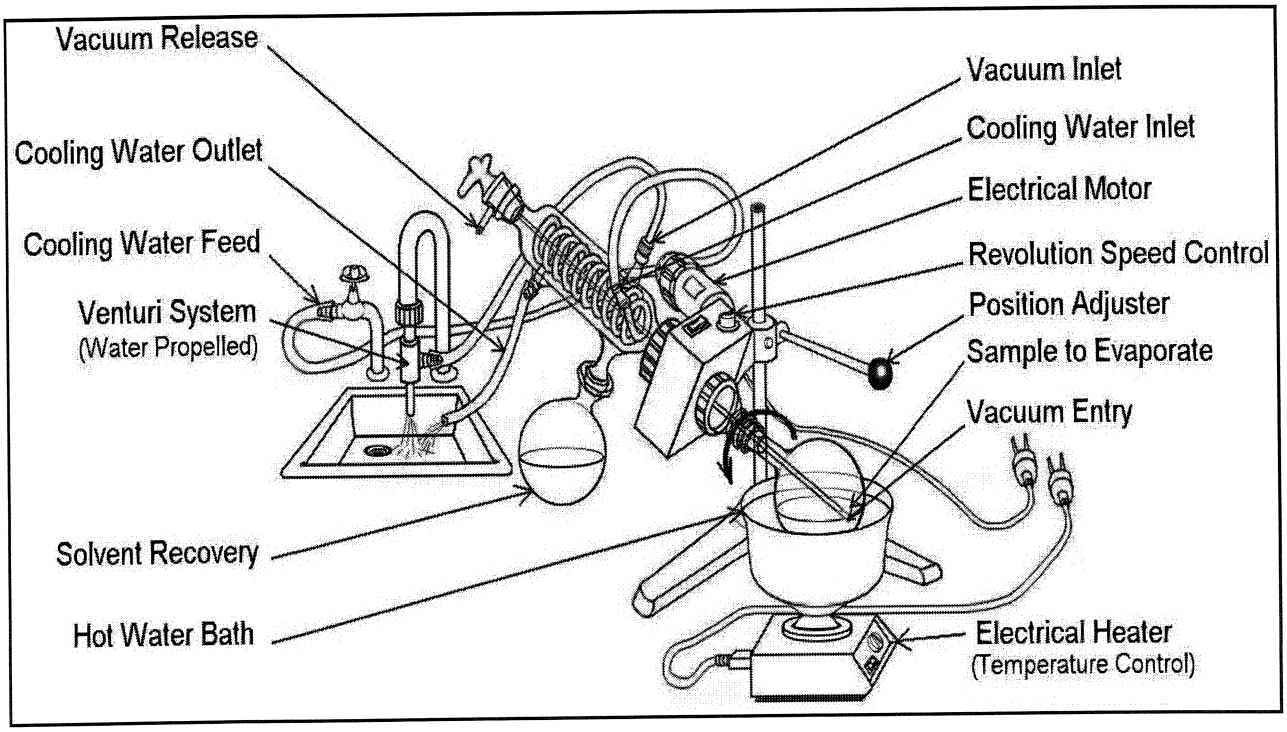


Figure 8: Description of the steam rotatory evaporator

In the next step, each concentrate of 10 ml was submitted to an evaporative process with a direct contact with a flow of nitrogen on the liquid surface, see Figure 9. The concentrate was evaporated until a volume of 2 ml. Great care should be taken to avoid looses by any emission of the solvent out of the recipient. The whole process takes approximately 8 minutes. The color of the concentrates is caused by the enrichment of the pollutants. At the beginning, the solvent is colorless and crystal appearance, and at

the end of the Soxhlet it acquires a yellowish color, and at the end of the steam evaporation the color was more deeply yellow, almost amber. At the end of the evaporation with nitrogen the color was almost brown.

The next step is to clean the samples from all sulphur and sulphur compounds because they severely interfere in the instrumental analysis. This step is carried out accordingly to EPA standard SS-42-35-217 to remove sulphur and sulphur compounds from the extract of a solid sample. First, 2 ml of isopropanol were added, 2 ml of TBA reagent (3.39 g of tetrabutyl ammonium sulphite is dissolved into 100 ml of nanopure water. After complete dissolution, washed with 20 ml x 3 times of *n*-heptane. The heptane was removed each time. At the end, the resulting solution was mixed with 25 g of sodium sulphite.) plus a spoon of sodium sulphite. Then the solution was shaken and warmed by hot water (50-60°C) and kept at that temperature during one minute. Five milliliters of nanopure water were added to the solution and shaken again. At the end, three phases or layers insoluble in each other with different colors could be seen, the upper cleaned organic layer return to the yellowish color and then separated and stored prior the gas chromatographic analysis.

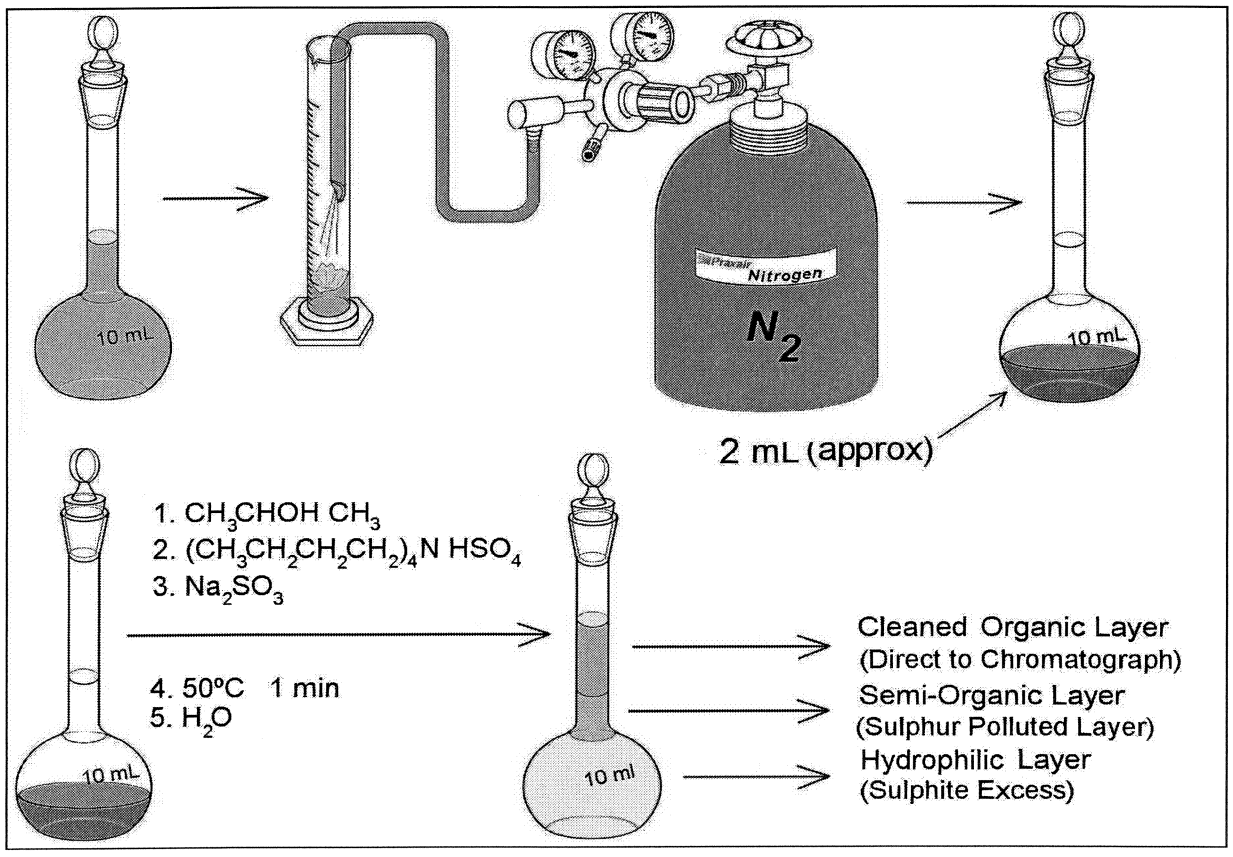


Figure 9: General procedure for sulphur removal and cleaning steps

3.3 Microwave digestion

Microwave digestion (MWD) has been used for three decades to get a more efficient extraction of metals in samples. The use of this method for organic compounds is quite new. It has been used for fiber headspace analysis of chlorobenzenes in soil (Santos *et al.*, 1999).

MWD was developed in order to accelerate the extraction rate. However, microwaves may effect the structure of organic compounds and care must be taken not to use microwaves of high intensity. The technique is influenced by pressure and temperature, so the process could be even faster. An important factor to consider is the use of more polar and environment friendly solvents. In some cases, water can be used. In this work a change in temperature and pressure in the microwave extraction was chosen (see Figure 10).

The procedure begins with the preparation of the sample already homogenized and weighed in the microwave container. This container is filled with 0.5 gram of the sample (sediment or soil) and the internal standard (50 µg of 0.5 ml of a solution of 100 µg / ml) is added as to the Soxhlet extraction samples. The container is made from very soft plastic (Nalgene, Teflon) and have a capacity of 100 ml. Then 10 ml of nanopure water was added to dissolve the solid sample. When the stopper is put at the top of this container, the sample with the water is shaken until it is homogenized. In this work 10 ml of sodium hydroxide solution (pH = 12 - 13, checked with pH paper) was added. This idea was implemented because of the acid character of the nonylphenols present and thought to be a help to make a more efficient extraction. The microwave equipment used in this work has the possibility to run 14 samples at a time in a circular revolver with one sample container used as temperature and pressure test and control. Each container has in a safety relief disc that will be broken in case of an abrupt pressure increase inside the container. The revolver with the 14 samples (tester included) is put into the oven (2 square feet approximated capacity) and the tester transductor is connected into the electronic receiver. The oven looks like a household microwave. The equipment offers the possibility to change with the pressure (0-50 psig) and temperature (30°-150°C). The revolver inside the oven rotate clockwise and vice versa just half revolution during the whole period of microwave exposition.

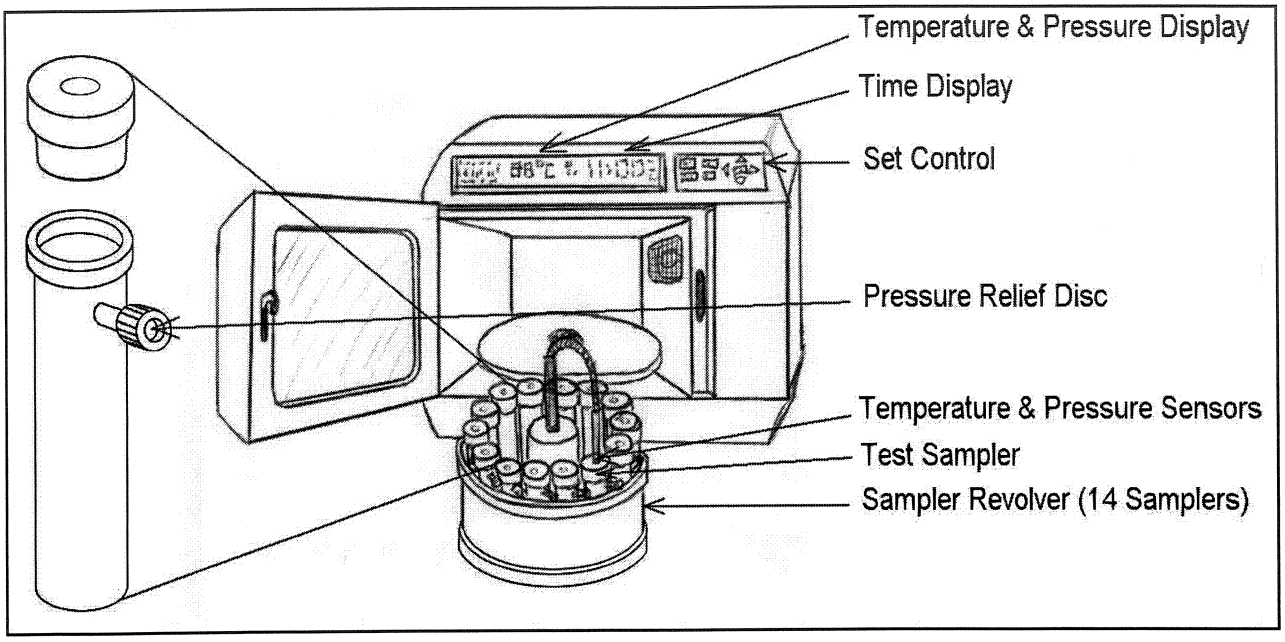


Figure 10: General scheme of the microwave equipment and samplers used

The samples were put in the microwave equipment with total exposure time of 50 min and a temperature of 80°C and a pressure of 30 psig. After the samples were submitted to this process they were left around 1 hour to get the normal temperature and pressure and let the solids precipitate again. Then the solid phase was removed from the samples, so they were filtered with standard filter paper in a glass funnel with the help of a vacuum system installed with a Kitta-Satto vessel and a line of 6 cm Hg of vacuum pressure (see Figure 11).

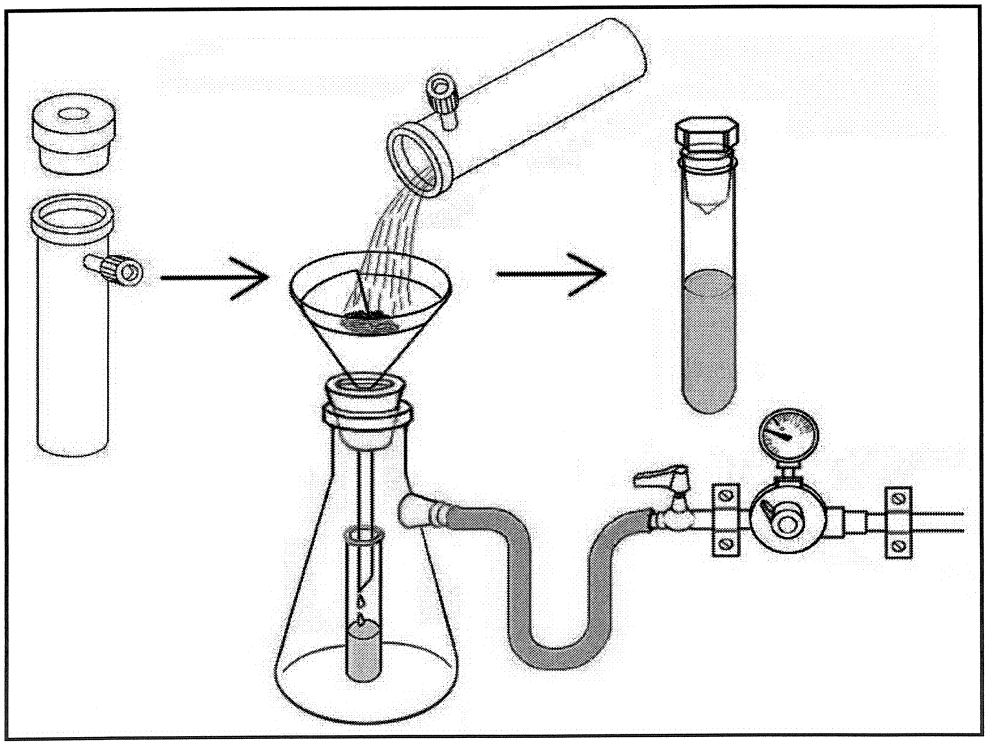


Figure 11: Filter procedure after Microwave digestion

3.4 Solid phase microextraction

The next step after the microwave extraction is to use the solid phase micro-extraction (SPME) to concentrate the organic pollutants in the aqueous sample and inject these organic compounds into a gas chromatograph. The sediment and soil samples were those that have been microwave digested, the water samples are just submitted directly to the SPME. This technique has improved the chromatographic analysis of organic pollutants in diluted aqueous.

The SPME technique is based on the principle of absorption/adsorption of organic compounds into a polymeric phase coated on a fused silica fibre. The procedure starts when a device similar to a syringe with a small diameter fibre on the top of the plunger, is put into contact with the aqueous sample solution (microwave digested extracts or just the water samples). In this case the fibre was kept for 10 minutes in the solution. It is important to use the same sampling time and the same agitation, since the absorption/adsorption process is governed by diffusion. The substance of interest will be sorbed into the polymeric phase on the fibre. This polymer phase used in this work was made of polydimethylsiloxane (PDMS, 7 μm).

This SPME procedure saves time compared with Soxhlet extraction (3 h ready to inject instead of more than 24 h) and one of the main objectives of this work is to compare the reliability of the results obtained with these two techniques. We took the results of the Soxhlet extraction as a base (supposing 100% efficiency), so the maximum concentration the MWD – SPME samples (only soil or sediment) could reach is the one obtained with Soxhlet extraction procedure.

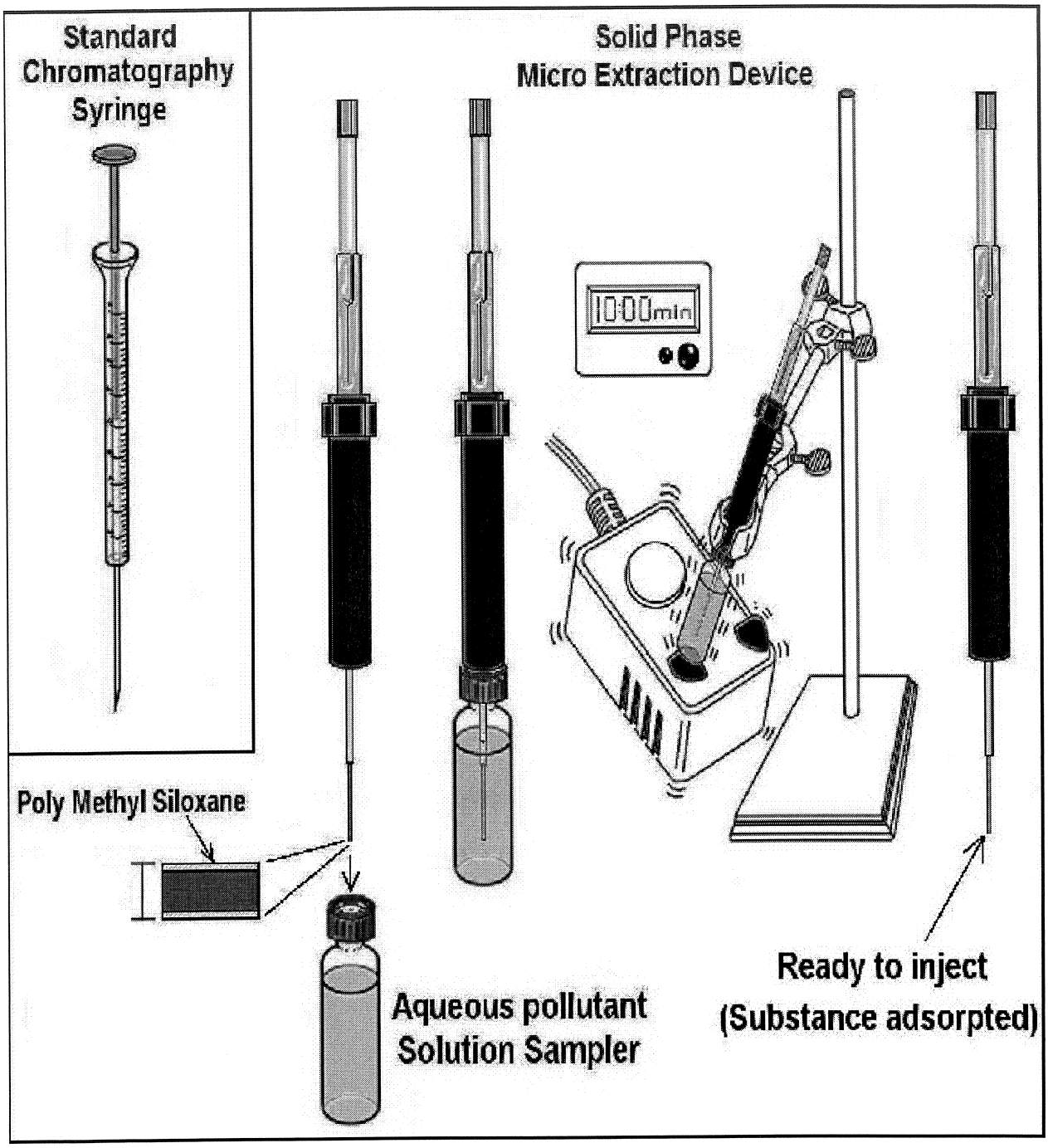


Figure 12: SPME device and stirring procedure used, compared with a standard chromatography syringe

3.5 Gas chromatography-FID and gas chromatography-MS

Today, many environmental analyses of organic compounds are carried out on gas chromatographic technique. A general scheme of the GC-FID equipment used in this project is shown in Figure 13.

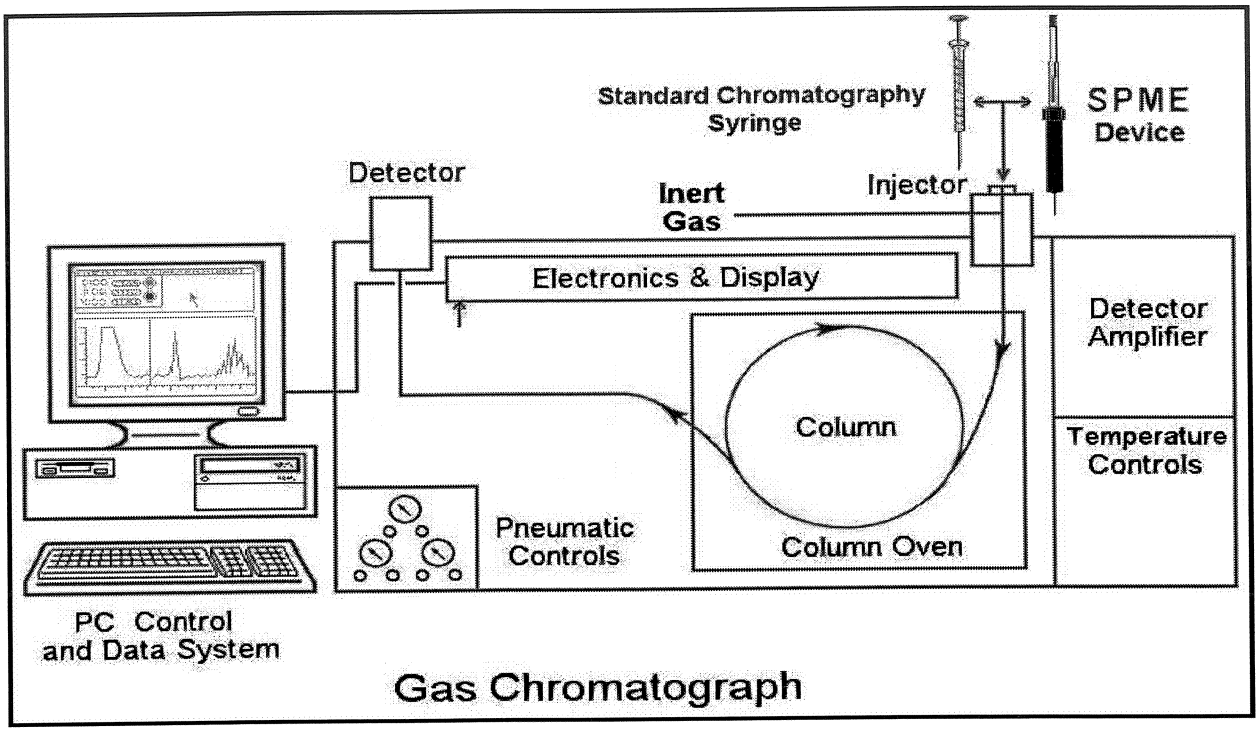


Figure 13: General scheme of a gas chromatograph equipment used

The volume injected of the Soxhlet extracts was 0.5 µl. A capillary GC column was used (stationary phase: DB5; film thickness: 0.25 µm; inner diameter: 0.20mm; length 30 m), see Figure 14. The running time for all the separations was 30 min, injector temperature: 260ºC, detector temperature: 250ºC, oven temperature: 90ºC, +7ºC / min to 260ºC.

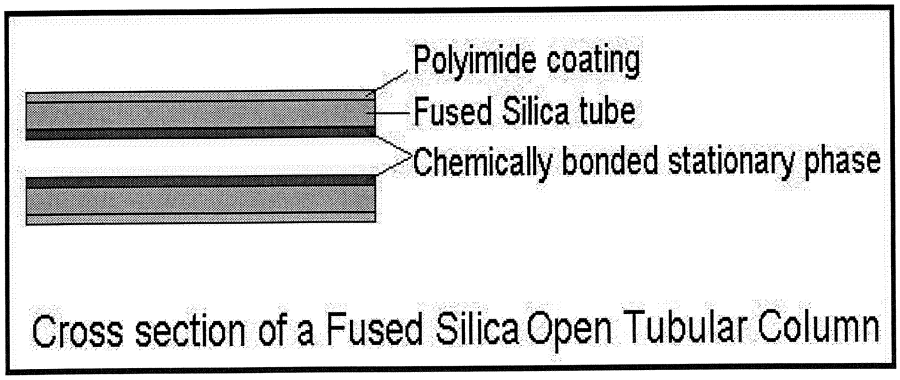


Figure 14: Cross section of the column

A flame ionization detector (FID) was used. FID is the most widely used detector for GC. It has a wide linear range ($\approx 10^7$). The response is roughly proportional to the total number of carbon atoms passing the detector per unit of time. For the principle of the detector see Figure 15.

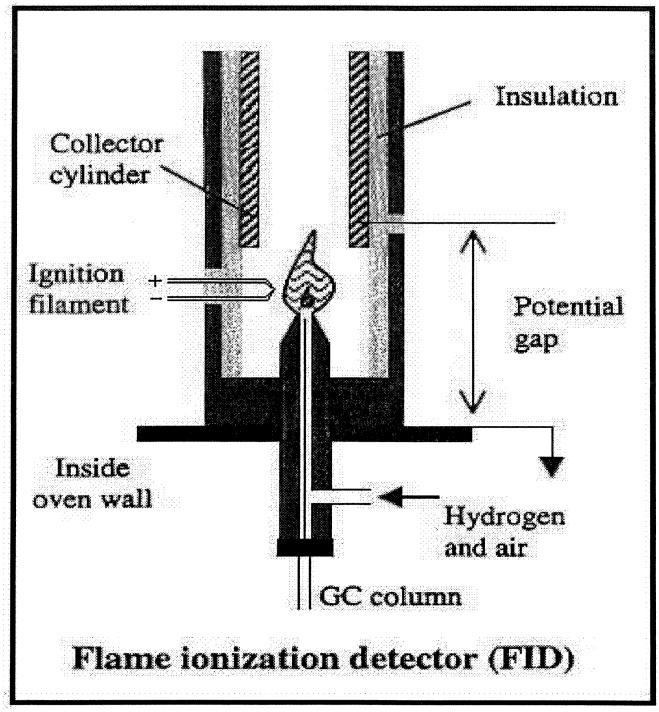


Figure 15: General scheme of the FID detector

For a few of the samples a GC-MS was used (see Figure 16). The mass spectrometer was a Varian Saturn Ion Trap 2000 coupled to the gas chromatograph Varian 3800.

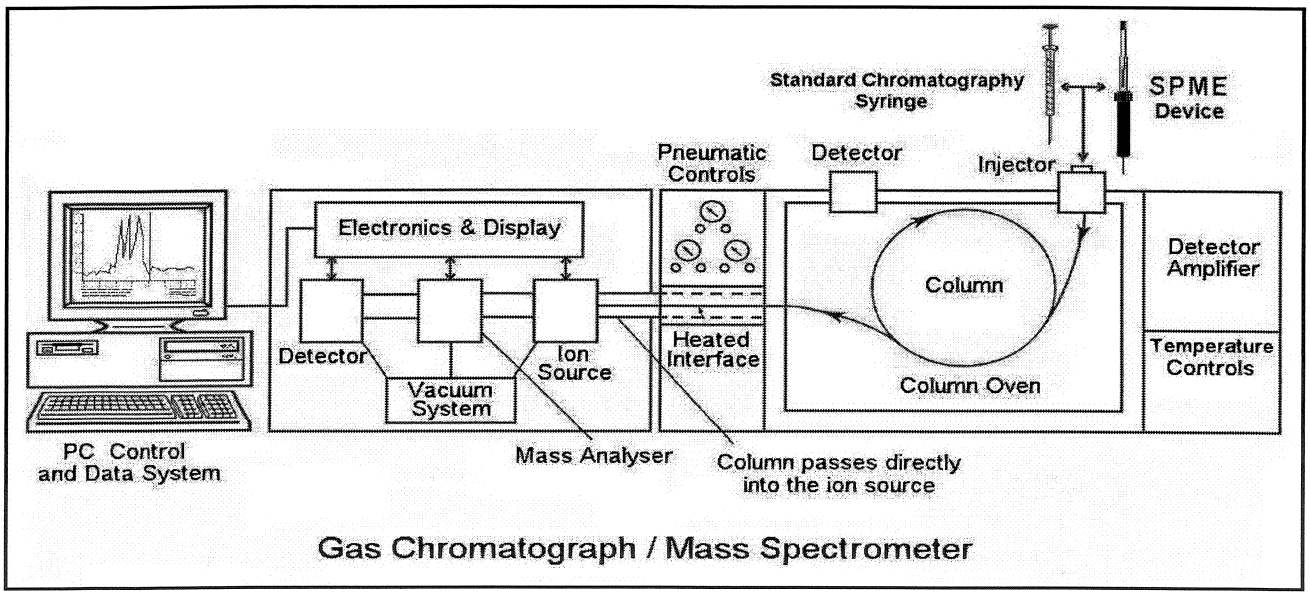


Figure 16: General scheme for the Gas Chromatograph- Mass Spectrometer equipment

The total running time was 54 min, with a *m/z* of 40-400 (sometimes only 135 mass number when searching for nonylphenol fragments) and 1 spectrum / s was taken. The initial oven temperature was 90°C and the final temperature of 260°C, and a temperature increase of 5°C / min. The injector temperature was 260°C. The injected volumes were the same as used in the GC (0.5 µl). A standard gas chromatographic syringe was used for the injection of the Soxhlet extracted samples and the SPME device for the microwave digested samples. The syringe was left in the injector until 10 min before the end of the analysis. The carrier gas flow was controlled by the pressure (22 psig) (see Figure 17).

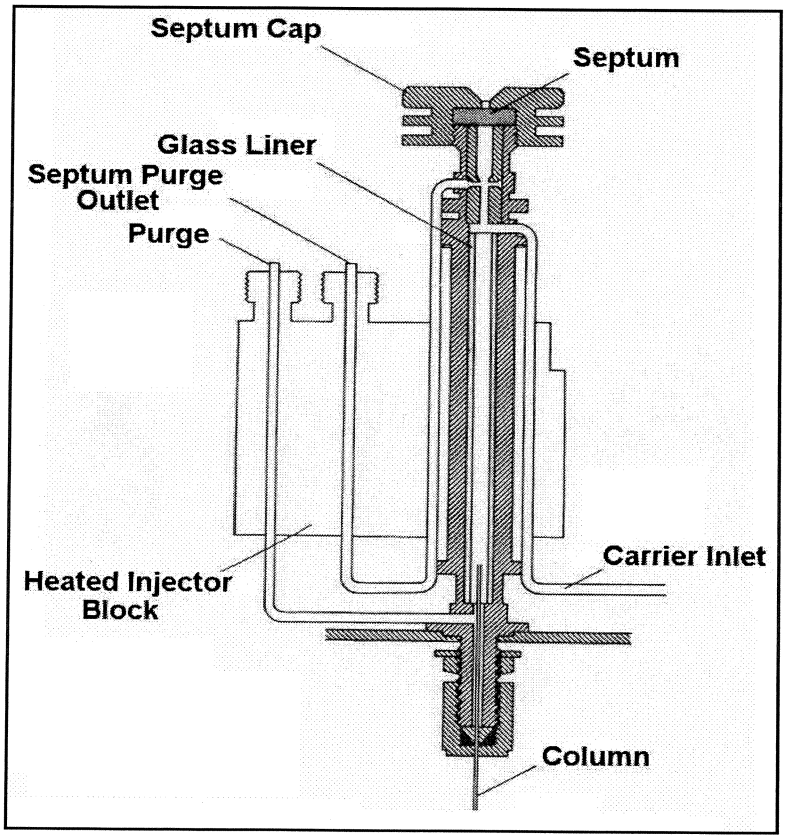


Figure 17: Split-Splitless vaporizing injector for capillary columns

CHAPTER 4:
ANALYTICAL METHODOLOGY

4.1 Weighing of samples

All the samples (soil and sediment = 22 samples) were submitted to a preliminary analysis where the dry weight and the organic content were measured. For these analyses, 5 g approx. was taken from each sample and weighed and kept in a crisol inside an electrical muffle at 110ºC for 24 h. The same crisol was put in an electrical furnace at 550ºC for 2 h. By these two techniques, the "dry weight" and "lost of ignition" was determined according to the Swedish Standard SS 02 81 13 (see references).

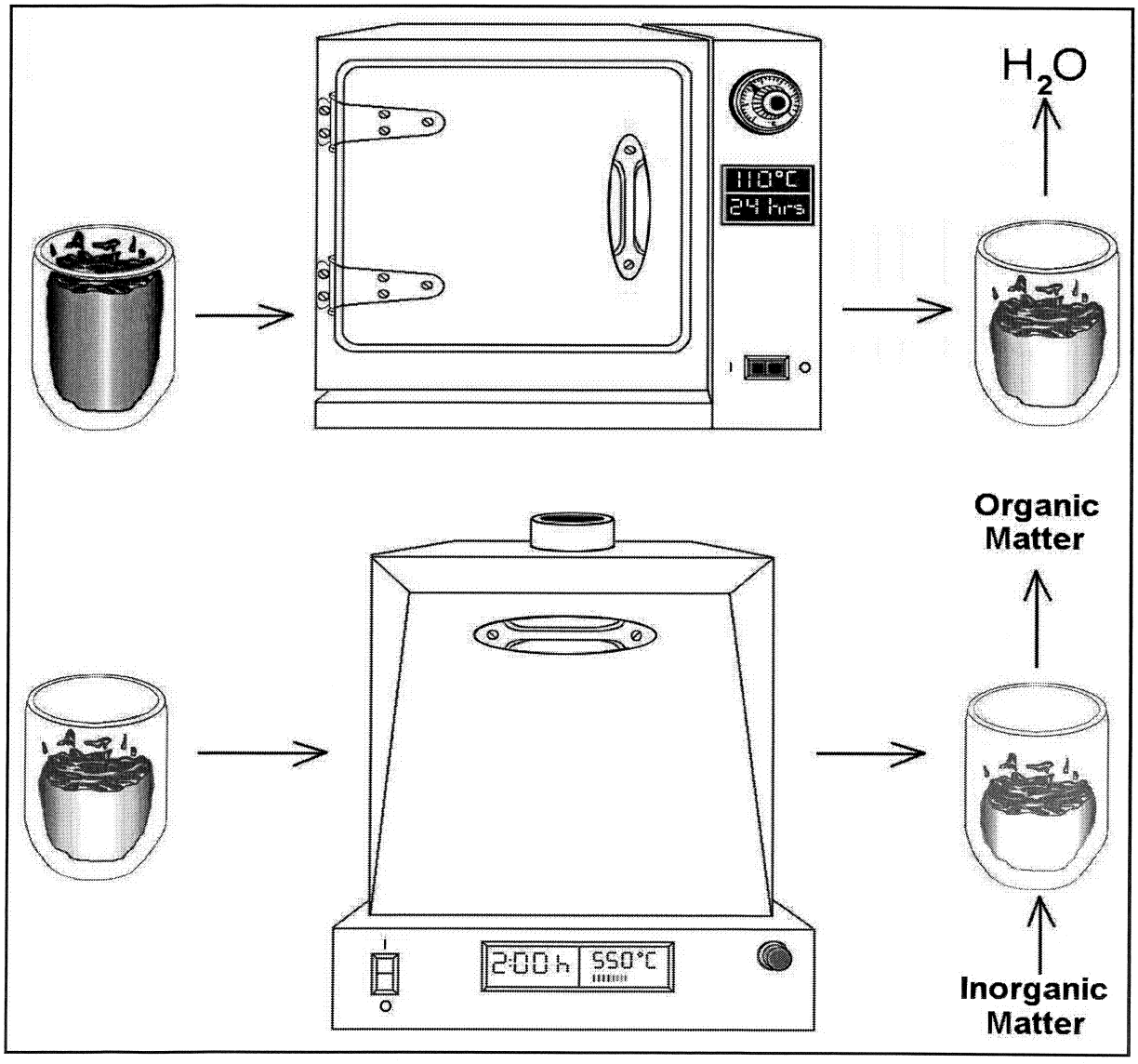


Figure 18: The techniques used to obtain the water, organic and inorganic content

Results and weights obtained with this analysis are shown in the Tables 1 – 8 in the Appendix.

4.2 Chromatographic results

The results from the chromatographic analysis gave three different chromatograms:

- All the chromatogram (with all peaks detected during the total running time).
- The zoom of the specific time range where internal standard* was found (only the IS peak is amplified).
- The zoom of the specific time range where nonylphenols was found or should be expected (only the family of peaks of NP is amplified)

From each analysis, a list of the peaks with its retention times and areas were obtained. The areas were used for the calculation of the pollutant concentration by comparison of calibration curves.

The complete graphs, areas and procedure for the most important samples and the internal standard injections (alone) are presented in the report. All chromatographic areas (soil + sediment with Soxhlet and MWD + water) are shown in the tables.

The internal standard peak was found within in a retention time range of 10.05 – 10.95 min in all the chromatograms. The retention time range of the nonylphenols was 16.65 – 17.95 min.

Representative chromatograms are presented in Figure 19-26 and peak areas in Table 3-5. The chromatogram from one of the most polluted samples (Soxhlet extracted) are shown in Figure 22 (sediment) and Figure 23 (soil). In these figures a brief description of the peaks, their size and distribution and meaning is given.

Concerning the techniques used for calibration see Appendix page 79 - 83.

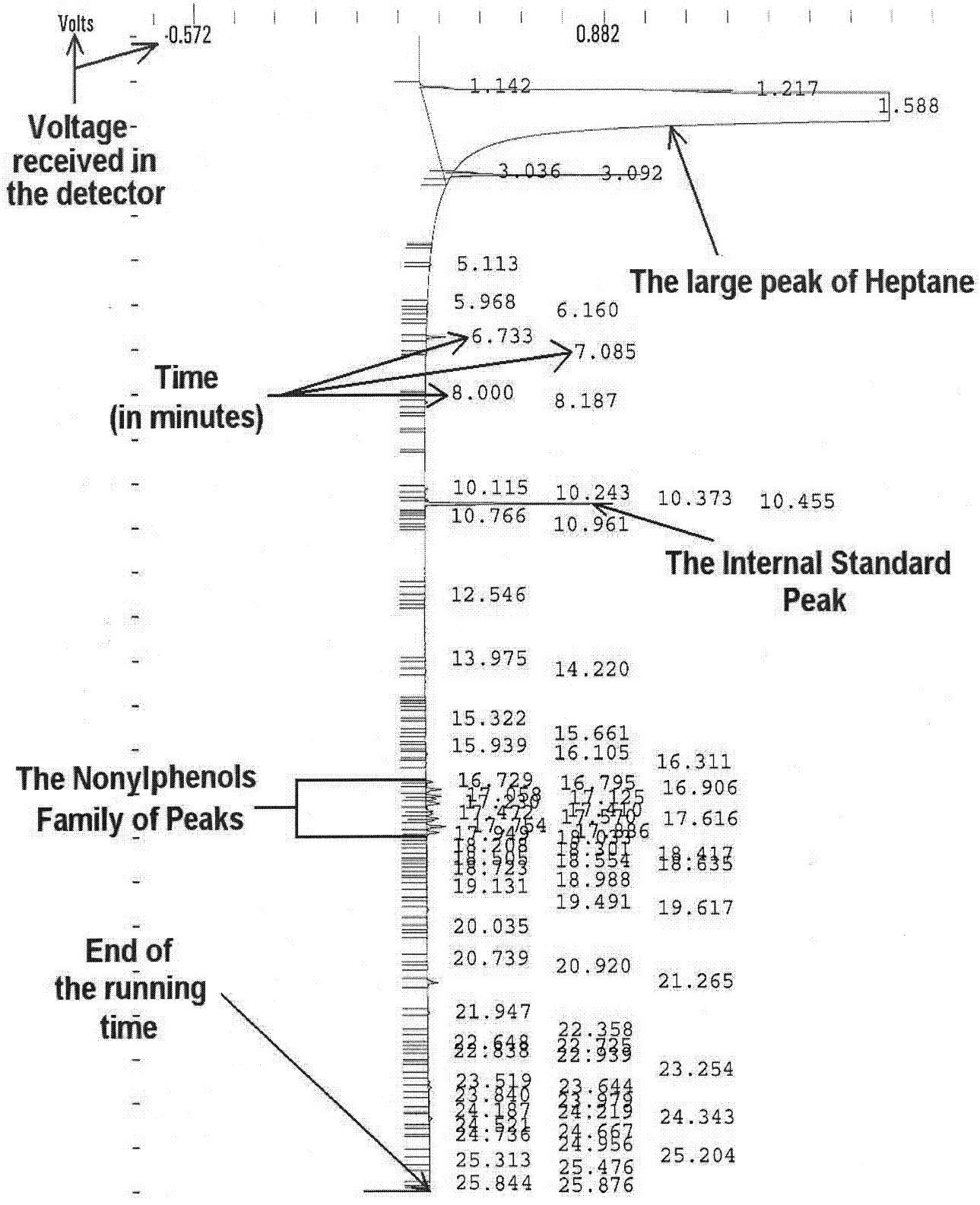


Figure 19: Chromatogram of the analysis of STANDARD I in heptane, internal standard 100 µg / ml and nonylphenols 25 µg / ml

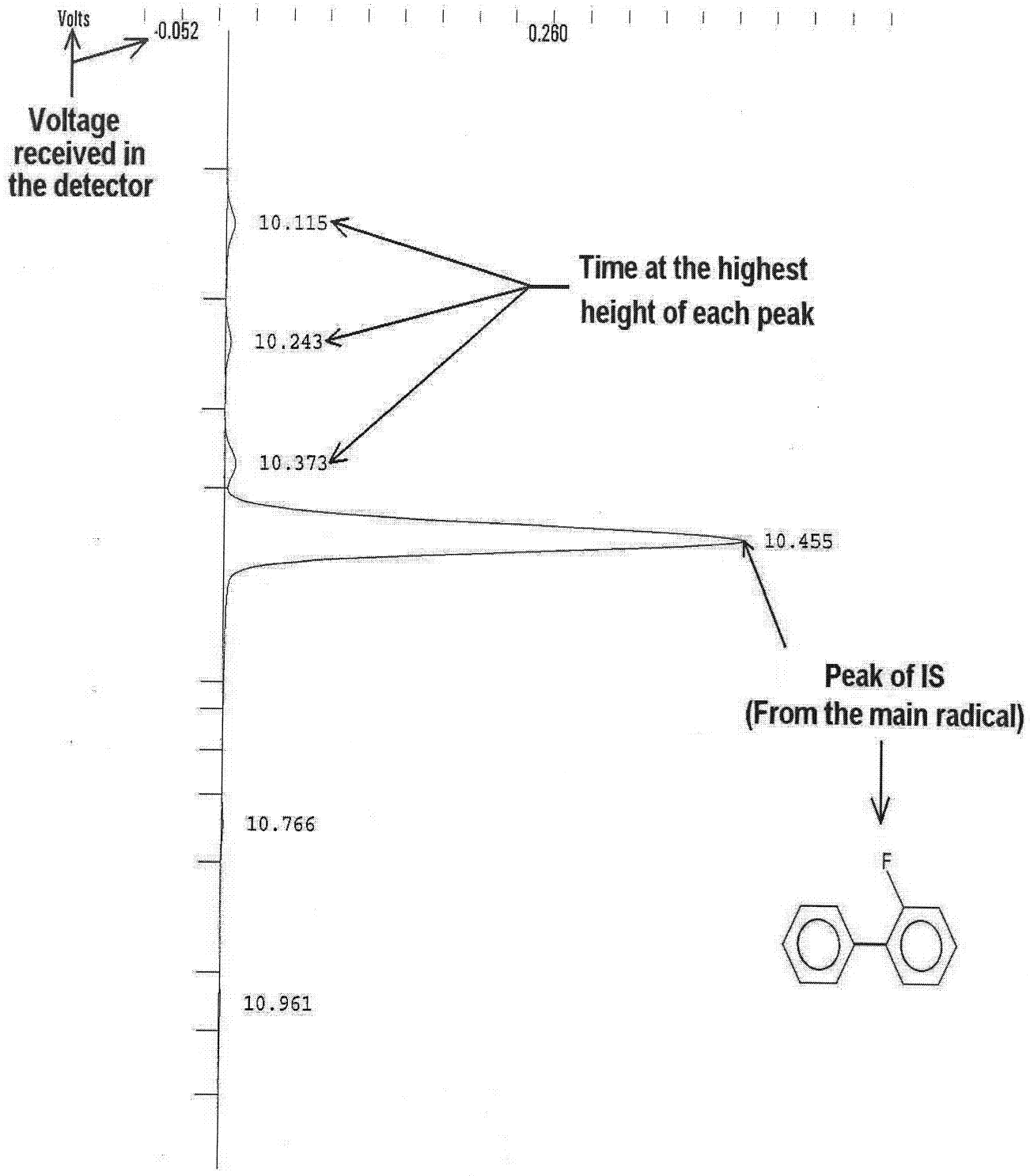


Figure 20: Chromatogram with the zoom of the internal standard area from Standard I

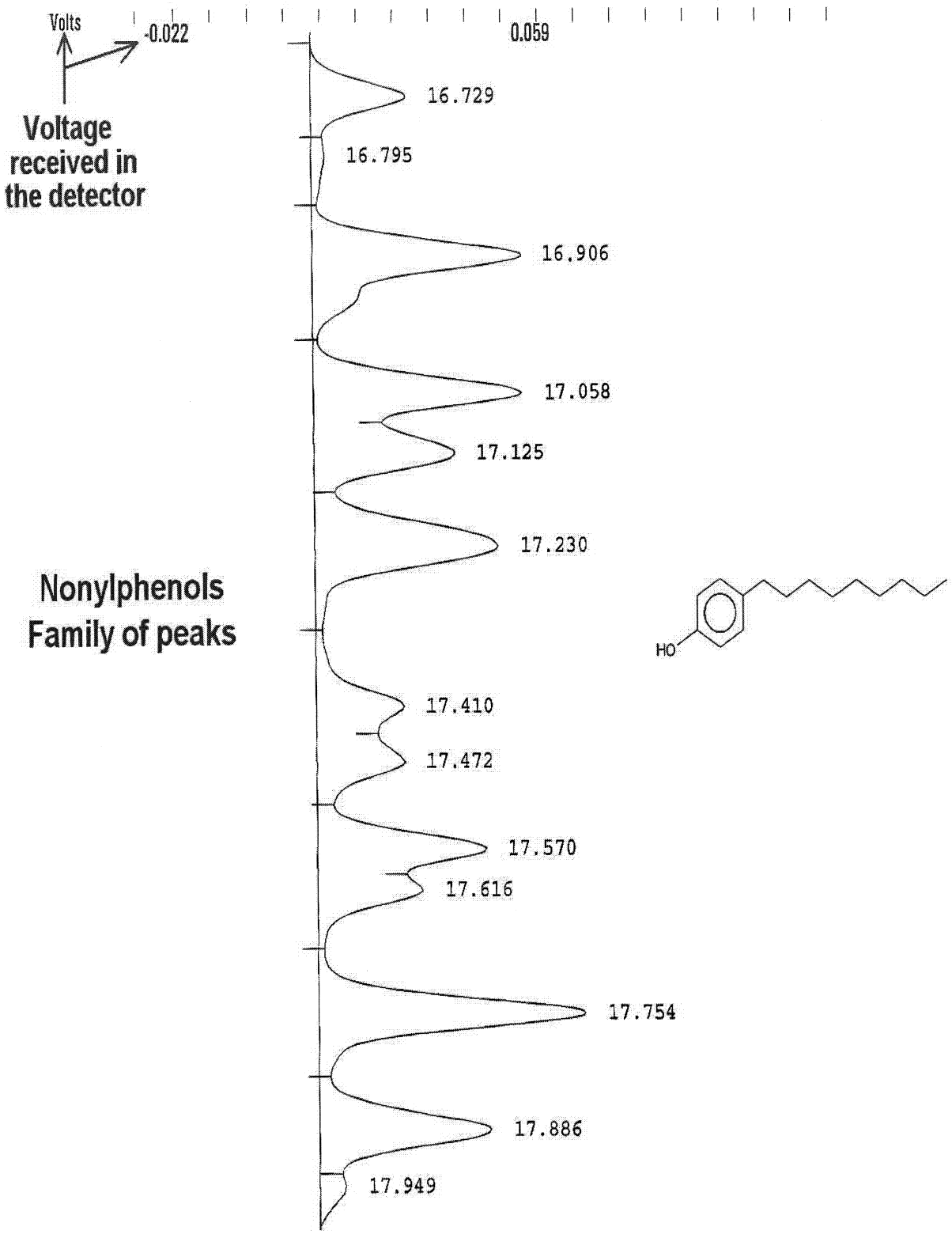


Figure 21: Chromatogram with zoom of the nonylphenols in Standard I

Table 3: Areas and retention times of each peak for the total chromatogram

Peak No.	Result ()	Ret. Time (min)	Area (counts)	Sep. Code	Width 1/2 (sec)
1	0.2134	1.142	121505	BV	1.9
2	3.9759	1.217	2263553	VV	3.0
3	89.5971	1.588	51009788	VB	47.6
4	0.1498	3.036	85277	TF	0.0
5	1.0230	3.092	582409	TF	0.0
6	0.0079	5.113	4470	BB	1.8
7	0.0037	5.968	2079	BB	1.8
8	0.0055	6.160	3134	BB	1.6
9	0.1400	6.733	79687	BB	1.8
10	0.0056	7.085	3160	BB	1.6
11	0.0039	8.000	2215	BB	2.2
12	0.0164	8.187	9334	BB	1.9
13	0.0221	10.115	12608	BP	2.0
14	0.0138	10.243	7835	PP	1.9
15	0.0302	10.373	17166	PV	2.1
16	1.3604	10.455	774500	VB	2.0
17	0.0021	10.766	1199	BB	1.6
18	0.0028	10.961	1603	BV	2.2
19	0.0031	12.546	1780	BB	2.4
20	0.0051	13.975	2891	BB	2.1
21	0.0103	14.220	5882	BB	2.3
22	0.0020	15.322	1111	BB	2.2
23	0.0036	15.661	2075	VB	2.2
24	0.0051	15.939	2898	PB	3.0
25	0.0263	16.105	14951	BB	2.6
26	0.0098	16.311	5599	BB	2.5
27	0.0653	16.729	37179	BV	2.2
28	0.0125	16.795	7111	VV	6.6
29	0.1763	16.906	100376	VV	2.4
30	0.1511	17.058	86004	VV	2.5
31	0.1176	17.125	66961	VV	3.3
32	0.1861	17.230	105934	VV	3.3
33	0.0786	17.410	44731	VV	2.7
34	0.0759	17.472	43209	VV	4.3
35	0.1255	17.570	71457	VV	2.6
36	0.0703	17.616	40024	VV	3.6
37	0.2009	17.754	114383	VV	2.4
38	0.1405	17.886	80011	VV	2.7
39	0.0172	17.949	9790	VV	4.6
40	0.0026	18.033	1464	VB	0.0
41	0.0061	18.208	3491	BV	2.9
42	0.0103	18.301	5866	VV	3.4
43	0.0106	18.417	6010	VV	3.0
44	0.0065	18.505	3687	VV	0.0
45	0.0071	18.554	4022	VV	3.2
46	0.0095	18.635	5411	VV	2.5
47	0.0063	18.723	3564	VV	2.8
48	0.0024	18.988	1377	BP	3.5
49	0.0061	19.131	3479	PB	3.1
50	0.0052	19.491	2975	VV	3.1
51	0.0205	19.617	11685	VB	2.4
52	0.0045	20.035	2540	BB	2.3
53	0.0044	20.739	2496	BB	2.5
54	0.0107	20.920	6093	BB	2.4
55	0.1219	21.265	69394	BB	2.3
56	0.0206	21.947	11740	BB	2.5
57	0.0019	22.358	1058	BB	2.8
58	0.0036	22.648	2041	BV	1.9
59	0.0037	22.725	2100	VV	3.2
60	0.0036	22.838	2032	VV	2.9
61	0.0074	22.939	4194	VP	7.7
62	0.0067	23.254	3817	BP	2.5
63	0.0259	23.519	14717	VV	3.2
64	0.0488	23.644	27790	VV	3.6
65	0.0138	23.840	7847	VV	0.0
66	0.0300	23.979	17106	VV	7.5
67	0.0080	24.187	4572	VV	0.0
68	0.0022	24.219	1255	VV	0.0
69	0.0583	24.343	33183	VV	3.0
70	0.0076	24.521	4350	VV	0.0
71	0.0157	24.667	8920	VV	0.0
72	0.0043	24.736	2459	VV	0.0
73	0.0305	24.956	17342	VV	0.0
74	0.0105	25.204	5983	VV	0.0
75	0.0096	25.313	5466	VV	0.0
76	0.0027	25.476	1534	VB	0.0
77	0.2955	25.844	168225	BV	4.5
78	0.2241	25.876	127608	VV	4.5
79	0.8740	25.969	497587	VB	8.0
Totals:	100.0004		56932359		

Peak times and Areas of the Internal Standard

Peak times and Areas of the Nonylphenols

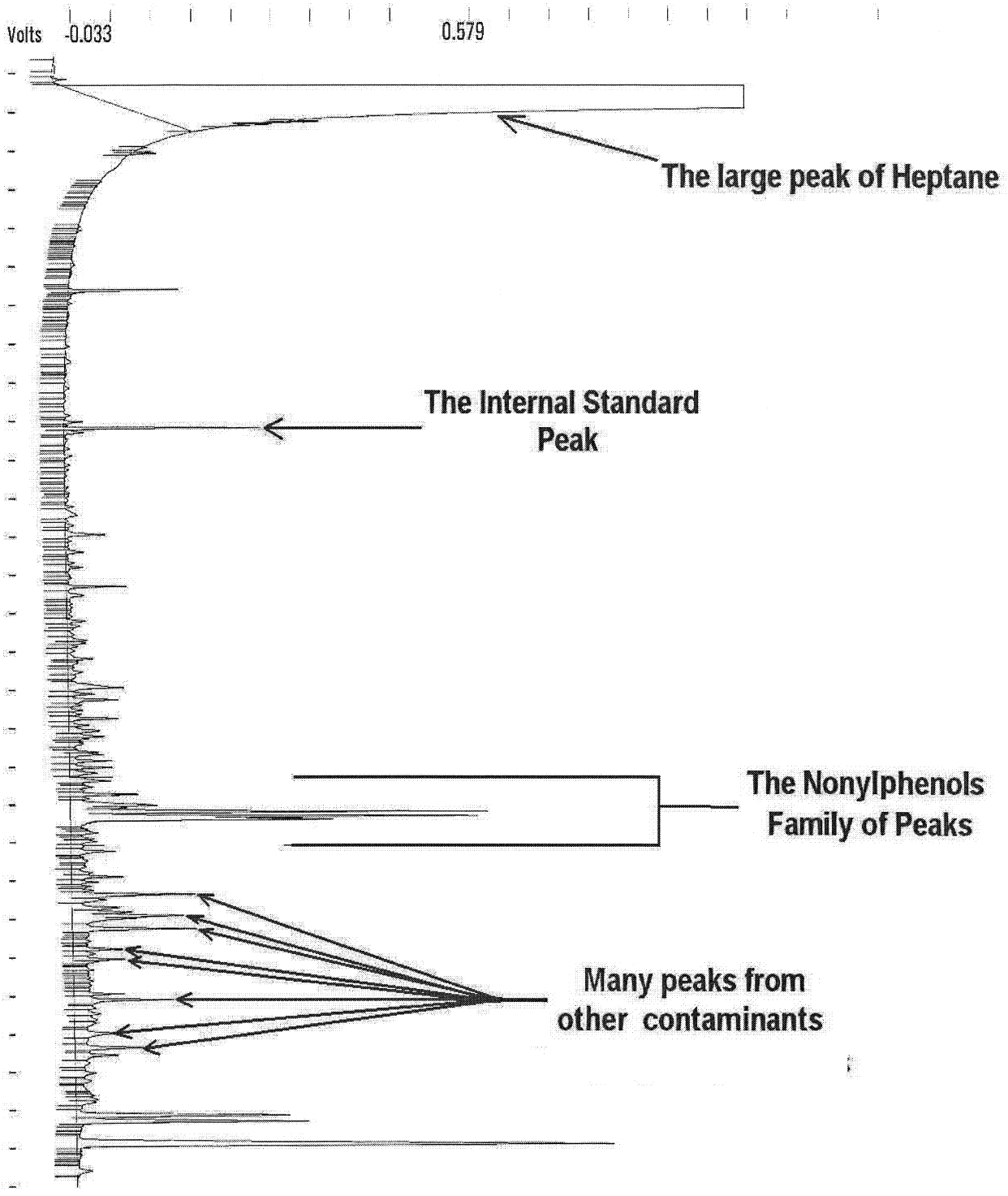


Figure 22: Total chromatogram of the sediment sample 11 extracted by Soxhlet and taken at ICA Maxi

Table 4: Retention times and areas of sediment sample 11 obtained by integration of the chromatogram

Peak No.	Result ()	Ret. Time (min)	Area (counts)	Sep. Code	Width 1/2 (sec)
1	0.0425	0.887	33599	BV	16.0
2	0.0371	0.946	29351	VP	17.3
3	0.0528	1.141	41727	PP	1.0
4	0.0062	1.262	4891	PP	1.3
5	54.5737	1.634	43166552	PB	53.7
6	0.0276	2.304	21867	TF	0.0
48	0.0536	9.818	42411	VV	2.5
49	0.0947	9.981	74938	VV	2.0
50	0.0148	10.075	11736	VV	1.7
51	0.8681	10.154	686636	VP	1.9
52	0.0869	10.260	68771	TS	0.0
53	0.0192	10.451	15189	TS	0.0
54	0.0328	10.683	25934	TS	0.0
55	0.0468	10.866	37012	PV	0.0
129	0.2727	19.150	215686	VV	6.3
130	0.3779	19.355	298945	VV	5.0
131	0.1053	19.465	83322	VV	0.0
132	0.1582	19.517	125131	VV	0.0
133	0.1104	19.618	87288	VV	0.0
134	0.4383	19.711	346680	VV	3.0
135	0.1593	19.822	126028	VV	0.0
136	0.9213	20.008	728740	VV	8.2
137	0.3438	20.067	271927	VV	0.0
138	2.5693	20.190	2032233	VV	3.0
139	2.3726	20.306	1876676	VV	3.0
140	2.1608	20.385	1709134	VV	4.5
141	0.0098	20.631	7740	TF	0.0
142	0.0069	20.684	5463	TF	0.0
143	0.1997	20.733	157957	VV	7.7
144	0.0524	20.833	41423	VV	0.0
145	0.1635	20.888	129359	VV	0.0

Peak times and Areas of the Internal Standard

Peak times and Areas of the Nonylphenols

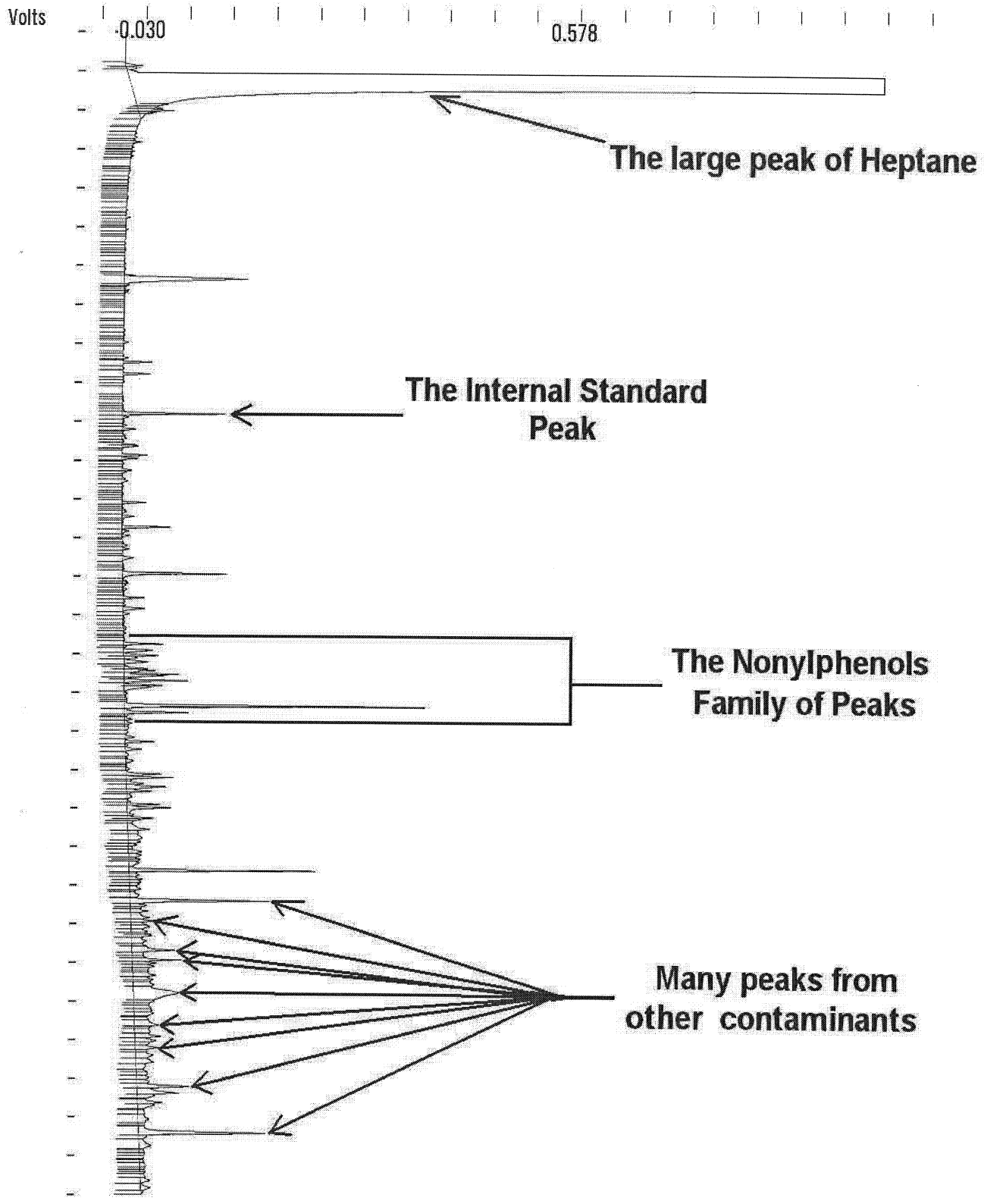


Figure 23: Total chromatogram from the soil sample 1 extracted by Soxhlet and taken at the southern part of the Akzo Nobel Mölndal plant

Table 5: Retention times and areas of soil sample 1 obtained by integration of the chromatogram

Peak No.	Result ()	Ret. Time (min)	Area counts)	Sep. Code	Width 1/2 (sec)
1	0.1085	0.847	51711	BV	2.5
2	0.0343	0.899	16335	VV	0.0
3	0.0796	0.919	37958	VV	3.6
4	47.8307	1.196	2804730	VV	0.0
5	13.7605	1.418	6560722	VB	4.1
53	0.0024	9.330	1168	BV	1.9
54	0.0031	9.411	1477	VV	4.5
55	0.0084	9.455	4028	VP	1.9
56	0.0055	9.564	2629	PV	2.0
57	0.0045	9.614	2163	VV	1.7
58	0.0111	9.694	5299	VV	2.0
59	0.6563	9.768	312907	VV	2.0
60	0.0303	9.856	14428	TS	0.0
61	0.0115	9.978	5463	TS	0.0
62	0.0218	10.069	10391	TS	0.0
63	0.0799	10.166	38099	VV	1.9
64	0.0081	10.217	3866	VV	3.1
123	0.0241	15.789	11481	VV	6.1
124	0.2638	15.886	125754	VV	3.6
125	0.2743	16.024	130778	VV	2.8
126	0.1193	16.085	56886	VV	5.4
127	0.2077	16.189	99030	VP	3.3
128	0.0288	16.313	13711	PV	4.7
129	0.1755	16.363	83658	VV	3.0
130	0.2417	16.413	115217	VV	3.1
131	0.3440	16.506	164021	VV	2.2
132	0.2343	16.544	111713	VV	3.0
133	0.0655	16.628	31211	VV	3.2
134	0.4218	16.672	201095	VV	2.2
135	0.2802	16.796	133571	VV	2.8
136	0.2439	16.838	116269	VV	5.8
137	0.0296	17.007	14123	VV	0.0
138	0.0094	17.091	4478	VV	0.0
139	0.0213	17.144	10178	VV	0.0
140	0.0193	17.190	9213	VV	0.0
141	2.0575	17.322	980952	VV	2.2
142	0.4387	17.492	209179	VV	2.3
143	0.0250	17.568	11902	VV	0.0
144	0.0062	17.606	2945	VV	0.0
145	0.0497	17.665	23689	VV	3.4
146	0.0679	17.770	32382	VV	0.0
147	0.0321	17.818	15313	VV	0.0
148	0.0464	17.895	22123	VV	0.0
149	0.0346	17.998	16515	VV	0.0
150	0.0812	18.131	38737	VV	3.9
151	0.2026	18.251	96612	VP	2.5
152	0.0245	18.345	11682	PV	0.0
153	0.1228	18.449	58543	VV	3.3

Peak times and Areas of the Internal Standard

Peak times and Areas of the Nonylphenols

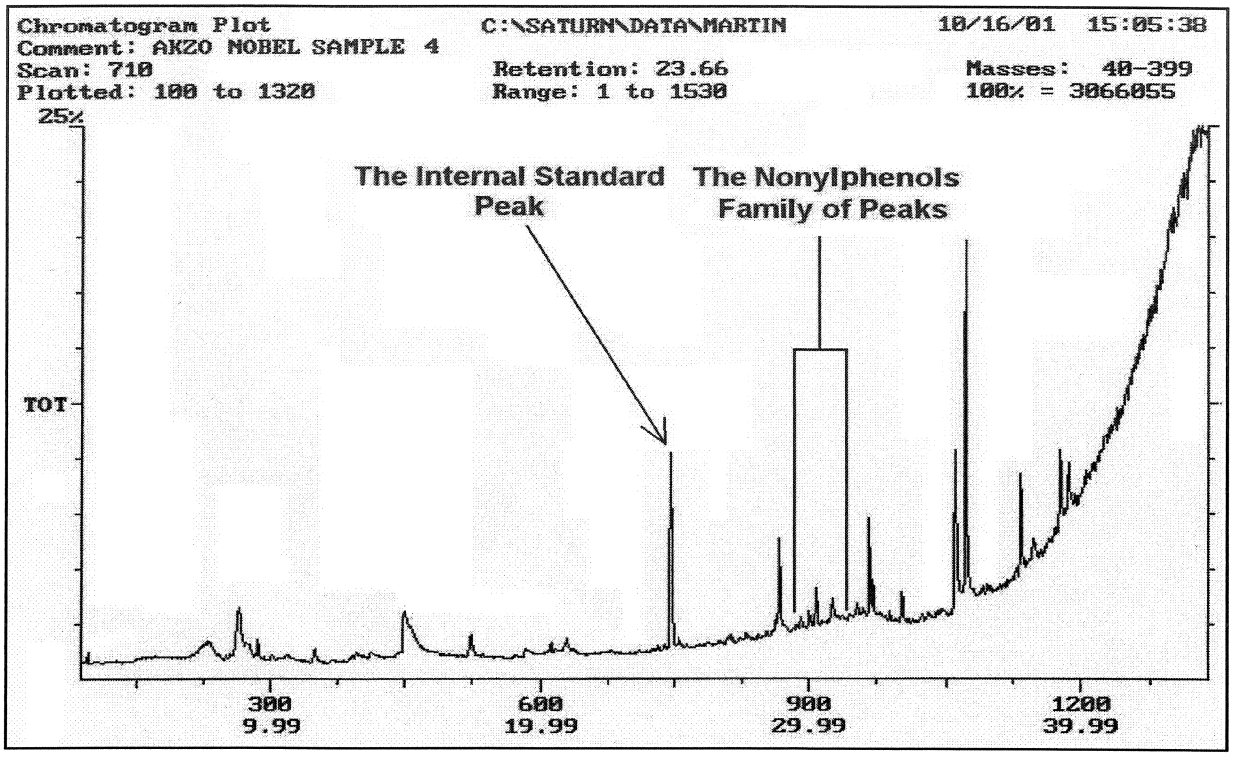


Figure 24: Total chromatograph of sediment sample 4 taken north of the Akzo Nobel Plant, Soxhlet extracted and analysed with the GC-MS equipment

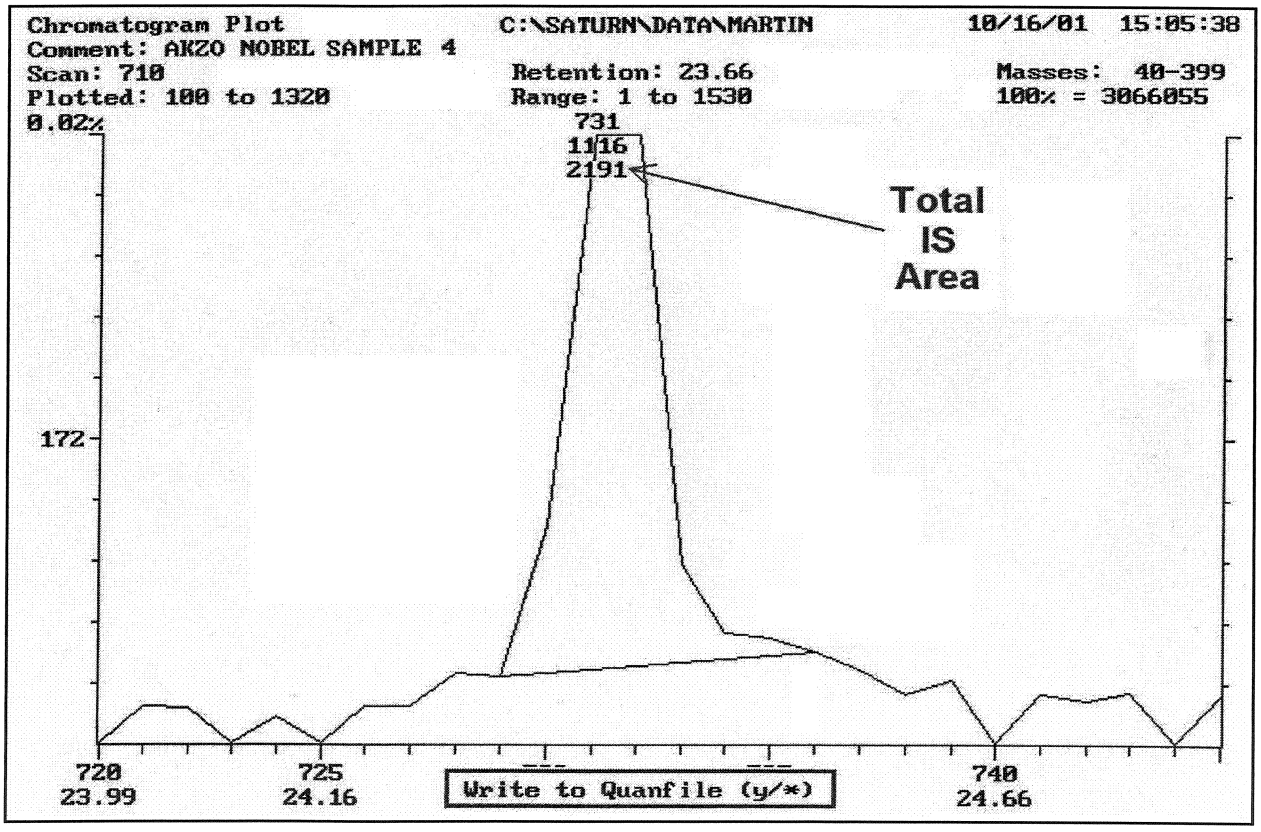


Figure 25: Zoom of the IS area of sediment sample 4 with the GC-MS equipment

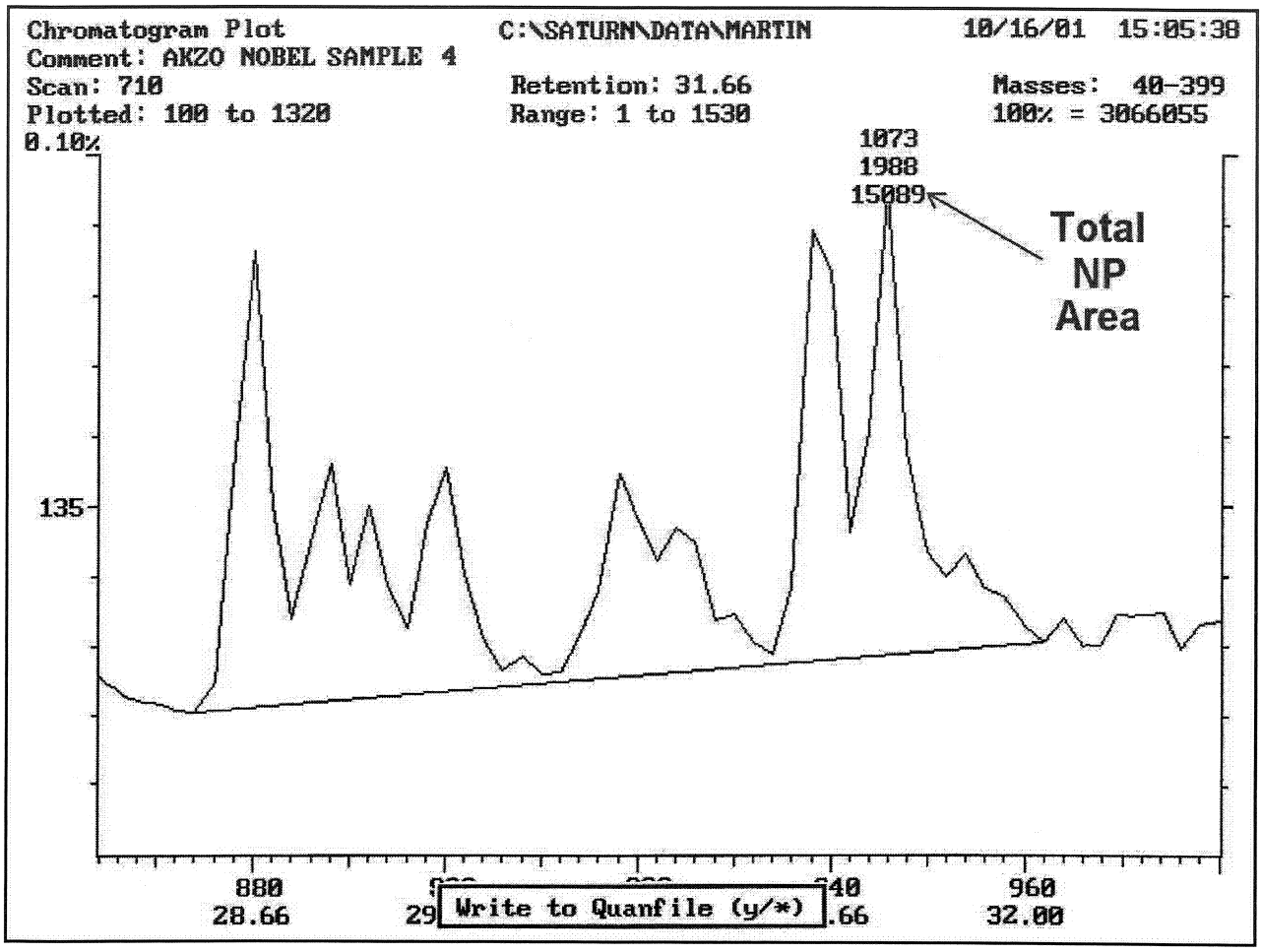


Figure 26: Zoom of the NP family peaks of sediment sample 4 by Soxhlet with GC-MS taken at the north of the Akzo Nobel Mölndal plant

The areas obtained by the integration of the zooms are:

A_{IS} = 2191
A_{NP} = 15189
A_{NP} / A_{IS} = 6.9324

The next step is to calculate the concentrations from the areas obtained in these Tables, but first we need to normalize and calibrate the use of the internal standard.

CHAPTER 5:
RESULT AND DISCUSSION

5.1 Concentration of nonylphenols

Table 6: Amount of nonylphenols / dry weight Soxhlet extracted sediment

Sediment Sample (SOXHLET) No.	[NP] (mg / kg)
1	65.9935
2	16.8677
3	13.3797
4*	78.0339
5	17.2572
6	55.0950
7	13.9045
8	18.2014
9	16.2101
10	9.0772
11	98.0363
12	16.8866

*This sample was submitted to GC-MS analysis

The concentrations of nonylphenols in the Soxhlet extracted sediment samples vary from 9 to almost 100 mg / kg dry weight. Even sediment samples taken at the north or south of the plant reach high values. This results show on a transport of nonylphenols to other locations, or indicate other sources of emissions of nonylphenols along the Mölndal river. Even samples that are far from the plant, especially No. 11 at ICA Maxi, reached the highest concentration found in any sample.

Table 7: Amount of nonylphenols / dry weight Soxhlet extracted soil sample

Soil Sample (SOXHLET) No.	[NP] (mg / kg)
1	68.5817
2	16.3475
3	16.0215
4	9.8694
5	26.9592
6	6.6176
7	62.0897
8	11.1132
9	7.5587
10	7.8899

In Table 7 the concentrations of nonylphenols in the soil at the former Akzo Nobel plant, outside the plant fence and close to the river, vary from 6 to 70 mg / kg and corresponds to a lower average values compared with those of the sediment samples. This indicates a massive transport, perhaps slow, from the soil to the sediment where nonylphenols seems to be accumulated. These concentrations could be a result of a slow degradation, transport and a possible transport to a deeper layer.

Table 8: Amount of nonylphenols / dry weight MWD-SPME water extracted sediment sample

Sediment Sample (MWD-SPME) No.	[NP] (mg / kg)
1	58.0821
2	16.5155
3	13.2657
4	10.1426
5	16.2180
6	48.6108
7	13.6885
8	17.5981
9	15.7856
10	8.7107
11	82.5508
12	16.5327
13	17.2738
14	11.7324
15	8.5666
16	45.7214
17	10.1342
18	24.3542
19	17.9266

In Table 8 the concentrations of nonylphenols in sediments samples extracted with water MWD-SPME are presented. The values vary from the Soxhlet extracted with a variance of only 2 – 3% and an average of 92% equal to the values reported in Table 6. These results shows are very good agreement between the MWD-SPME and the Soxhlet extraction method.

Table 9: Amount of nonylphenols / dry weight MWD-SPME water extracted soil sample

Soil Sample (MWD-SPME) No.	[NP] (mg / kg)
1	64.9590
2	15.5918
3	14.9658
4	9.5800
5	25.7409
6	6.4579
7	60.1900
8	10.7209
9	7.5062
10	7.7527

In Table 9 the values of the soil concentration are also almost the same as those obtained from the Soxhlet extraction, and strengthen the reliability of the MWD-SPME water extraction used in this project.

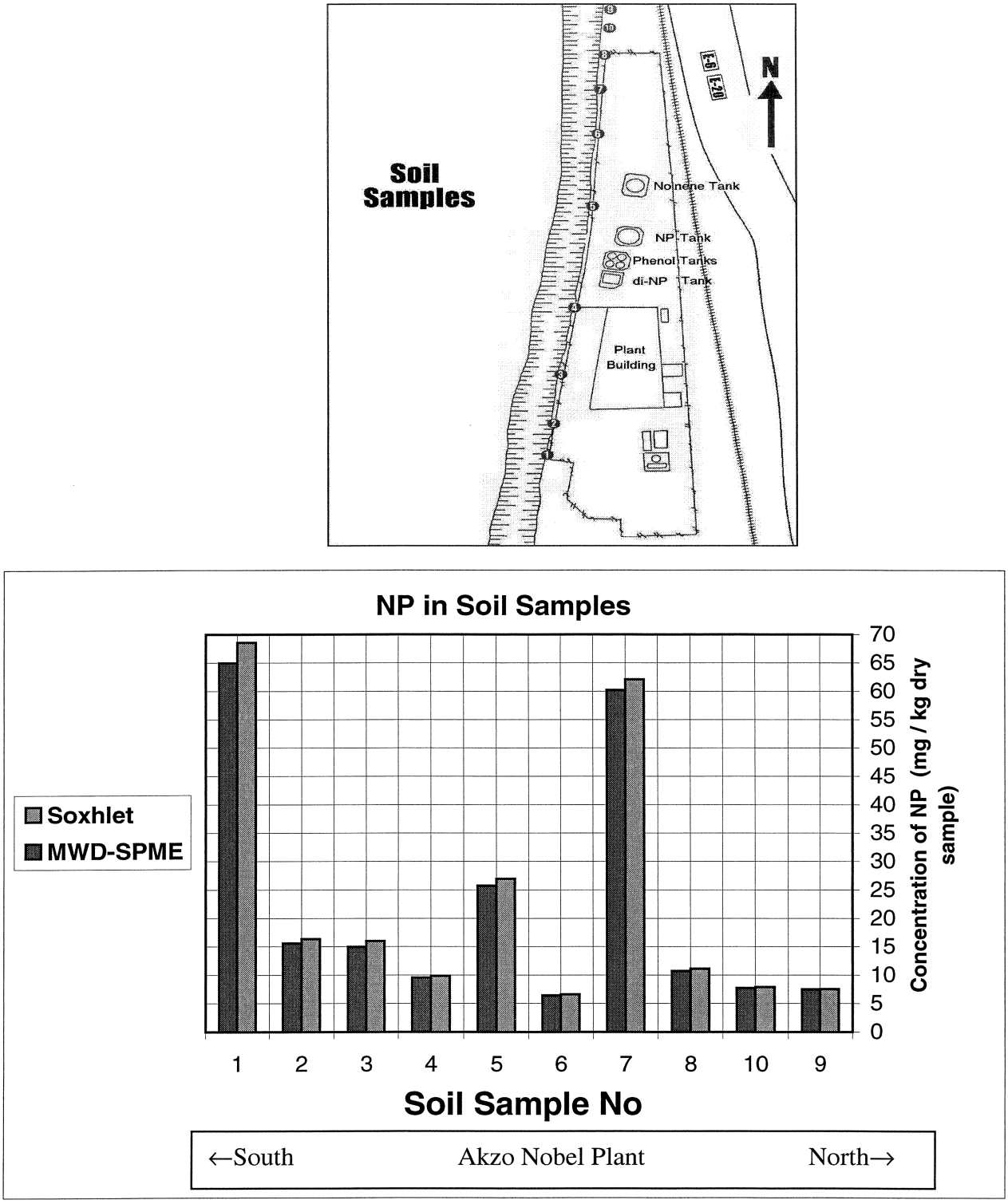


Figure 27: Comparison of results obtained with Soxhlet and MWD-SPME in soil samples

In Figure 27 a comparison of the amounts of nonylphenols in the Soxhlet and MWD soil samples is shown. The sampling positions at the Akzo Nobel Mölndal plant are also shown in a map. The highest concentration was obtained at the southern part of the plant, which suggest the importance of further studies of the surrounding areas, both down- and upstream the Mölndal river and in depth at the factory area to get depth profiles.

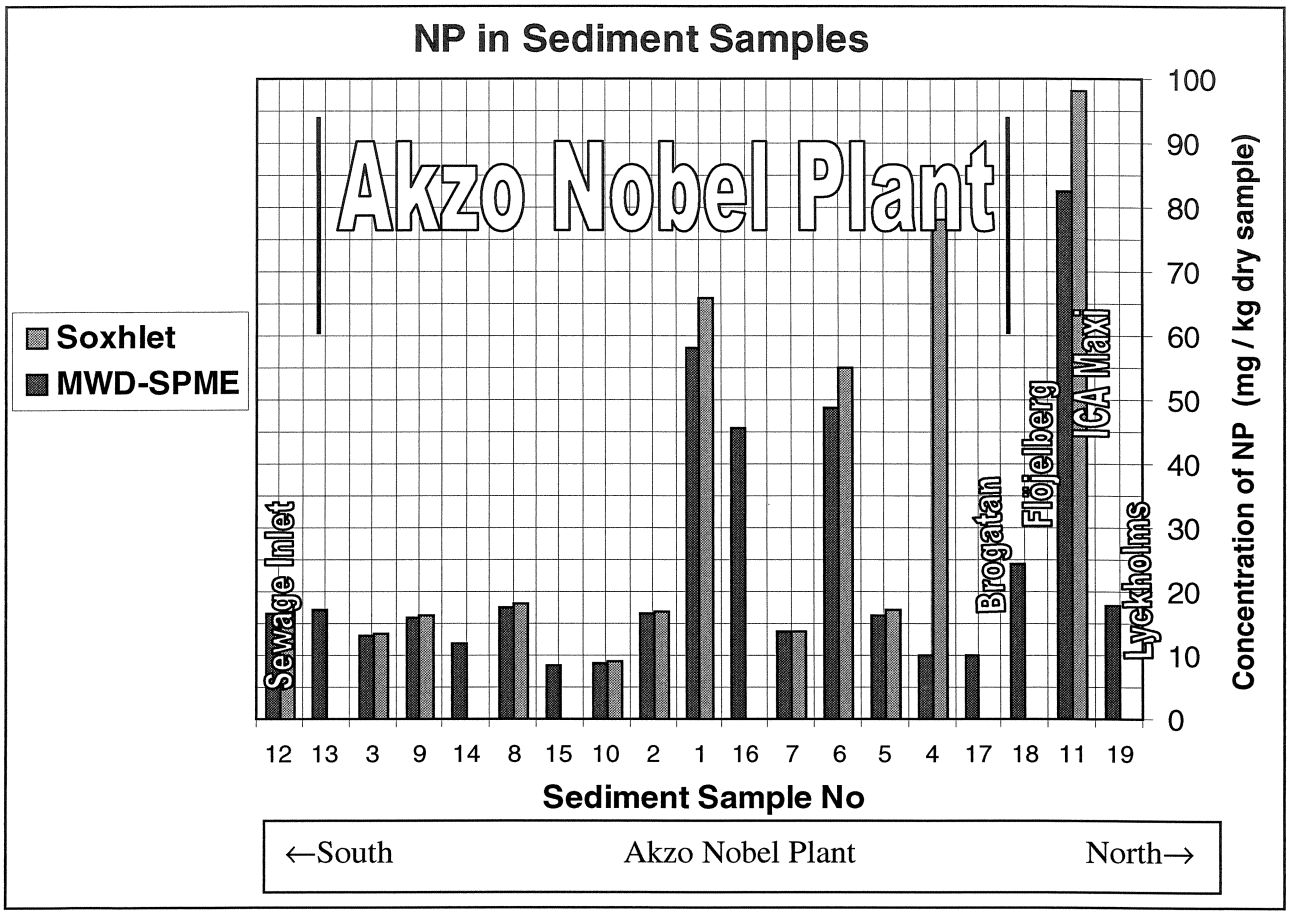
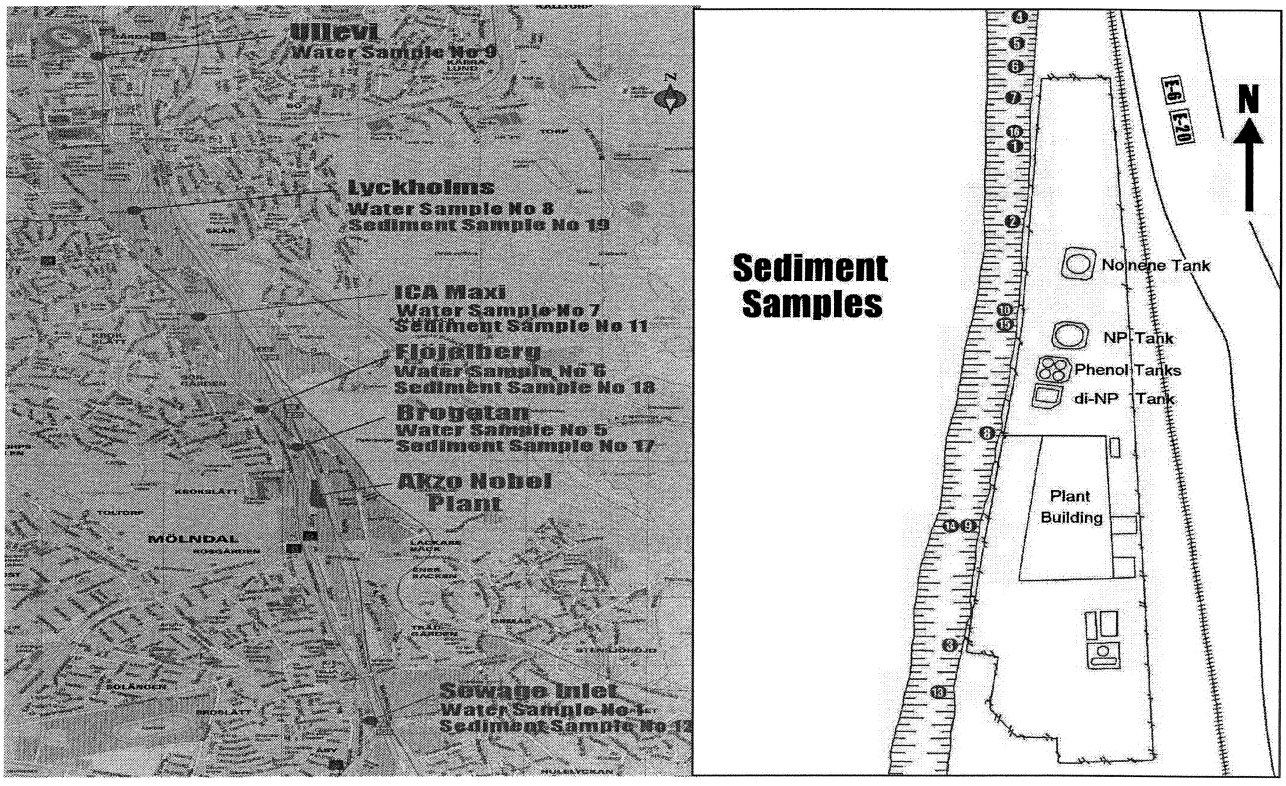


Figure 28: Comparison of results obtained with Soxhlet and MWD-SPME in sediment samples



In Figure 28 a comparison between the Soxhlet and MWD extracted sediment samples is presented. The values are similar in both cases and indicate a good agreement between the two methods. The results also show that the concentration vary at the locations studied and suggest further studies both up- and downstream the factory area until the values obtained reach zero and indicate where the transport could stop. Analyses of the Göta river sediment are also suggested to investigate if any nonylphenols are present.

5.2 Discussion of the results

From the results presented in this study it is obvious that the sediment had higher concentrations (10 - 98 mg / kg) of nonylphenols than the soil close to the river at the factory site (6.4 - 69 mg / kg). The result obtained varies from a few mg to almost one hundred mg per kg of soil or sediment. The concentrations in all the water samples analysed were too low for detection. The sediment and soil are highly polluted and immediate remediation should be carried out. The high concentration found in the sediment sample up stream the plant, at Mölndalsbro sample 12 – 16.5 mg / kg, suggest that the pollutant has been transported several kilometres by some mechanism. The nonylphenols at this site could be explained by a continuous transport of contaminated sediments towards the stream of the river, or accidentally sediment transports when the river have flooded several times during the last ten years. The high concentrations of nonylphenols at Mölndalsbro could also indicate other possible sources of emissions along the river.

The most superficial layer of soils (a very thin layer = 10 -15 cm) presents a high leaching process which transports almost every organic pollutant in a small period of time because of their physical structure and acidity. The transport in the deeper layers is very slow in any direction, especially when the season is cold and the water inside these matrices is frozen and makes the transport through other matrices almost impossible. With this assumption it is clear that higher concentrations could be found in a deeper layer in the soil, and further analysis of the soil samples through the depth should be carried out at least until 1 m depth, especially when a high concentration is found in the surface layers.

The theories about transport mechanisms suggest a probable mechanism of transport where the pollutant is originated in the soil matrix, going to the water and sediment through surface contact in an accumulative process that start from the water to the sediment, where it prefer to stay for a certain period of time. This corresponds to the values of concentration obtained in this work, but should be verified with further studies (more sampling) and a time profile (analysis of samples taken in a progressive period of time), and understanding the possible degradation which occurs naturally. For time reasons, the probable transport mechanism and fate of the nonylphenols in soil, water and sediment need to be further studied.

5.3 Possible scenarios

Earlier works presented (Payá-Pérez *et al.*, 1992) reveals that the fate of a very non-polar organic substance will be dominant in a solid phase rather than an aqueous phase. This is explained by the partition coefficients for the substances, when the compound reach the equilibrium between two phases, it tend to accumulate in the pores and surfaces of the solid, and will only be present in very low concentrations in polar solvents. The spodosol type of soil distinguishes from other soils in its high acidity (making the transport of a substance which ionises at low pH easier), easy leaching transport in its first layer (from where the pollutant originates) and a secondary or deeper layer that normally stops the transport of the substances through the deepest layers. Thus, a maximum concentration of nonylphenols could be found at a certain depth. Taking this fact as possible, the pollutant has only two ways to move: suffer an oxidative and biological degradation or move to any other matrix. In order to check the biodegradation and natural removal of this substance in the soil, it is necessary to carry out a time sampling analysis where the concentration of the pollutant is determined during a long period of time with the same frequency, place and depth. This could prove the fact that the pollutant has to move to the neighbouring medias. The closest media is the water of the river, which could present the contact in the most superficial layers of soil rather than its sediment, but the water possess a very high transport of the possible solvated pollutants found there. The other phase in indirect contact with soil is the sediment of the river. A sediment and a soil with a high organic content (TOC) has a high ability to adsorb and accumulate organic pollutants. Lost of ignition measured in this project could be used as an indication of high or low organic content of soil and sediment samples.

The possible original scenario, a non-polluted soil in contact with a continuous emissions and accidental spills of nonylphenols only from the factory near the river. When the contact ended the soil presented the higher concentration in areas extremely close to the plant if the transport and degradation were low. But the results obtained in this work indicates even the higher concentrations are reached very far from the plant and in the sediment, not in the soil. This indicates a possible short retention of the pollutant in the soil and a transport mainly to the water and in a smaller amount directly to the sediment. So in some period of time (less than the closure of the plant) the concentration in the soil was maximum but was kept at this level during the rest of the operational time of the plant. The water reached the highest concentration very fast (perhaps almost zero concentration) but the high transport it presents made the pollutant reached far distances from the plant (up- and downstream). The sediment started to accumulate the pollutant in its matrix, and the concentrations obtained began to reach a level almost equal to the soil (32 mg / kg average) thereafter, an accumulative effect made it to reach even higher concentrations than the soil (23 mg / kg average). A transport of polluted sediments along the river is also a probable transport process for the nonylphenols from the factory site.

5.4 Fate of pollutants

The final fate of nonylphenols in soil, sediment and water is influenced by many environmental factors like the contribution of the global warming and the change in the weather. This change could involve an increase of the temperature, which produces a higher leaching and transport of the pollutant through the soil to the water and sediment, and a higher leaching from the superficial soil to deeper stratus (more difficult to be removed) and a less amount of oxygen available in these three medias for an oxidative or biological degradation of nonylphenols.

The global warming also affects the pH in the environment, which could involve a faster transport and removal of the pollutant from the soil. Global warming also increases the capability to dissolve more organic matter into the water, which results in a higher capacity of the pollutant to reach further distances and ecosystems. Another important factor to study is the capacity of the sediment to accumulate the pollutant and the biological and geophysical ability to degrade it. Other possible effects of the accumulation of nonylphenols in the sediment is the continuous equilibrium between it and the water that could affect any animal and plant living in this ecosystem by chronic, subchronic, acute or bioaccumulative effects. The toxic characteristics of nonylphenols are discussed later (Scott *et al.*, 2000).

A possible scenario of the transport of the nonylphenols along the Mölndal river, involves a slow transport from the soil to the sediment and water, and the concentration in the soil will be high for a long period of time without any remediation. The water will continue increase its concentration of nonylphenols in a level which could make the pollutant reach areas far away areas and could cause damage to other ecosystems (see Figure 29). The polluted sediment could damage living organisms and bioaccumulative effects could be present. As the highest sources of pollution have been closed down, the levels of nonylphenols in the sediment are probably going to increase until some period of time and only then a reliable study of the degradation of this pollutant in the soil could be measured (Depledge *et al.*, 1999)

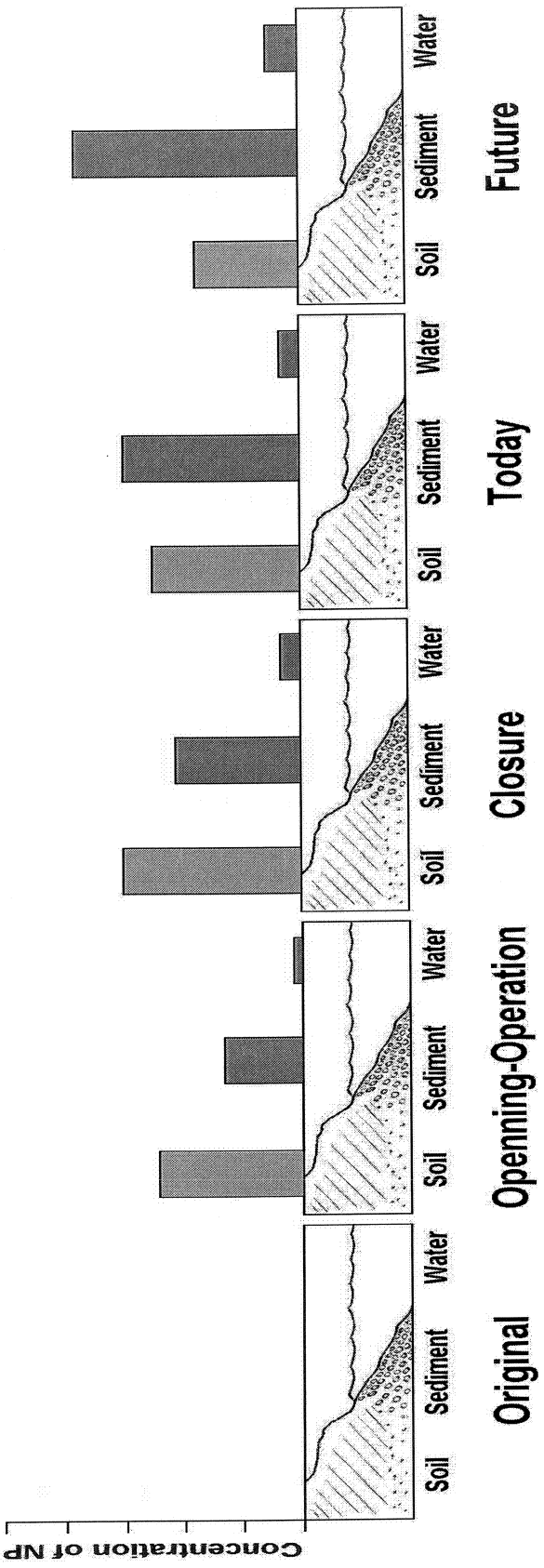


Figure 29: Scenarios of the concentration of nonylphenols in sediment, water and soil at the former Akzo Nobel Mölndal plant

CHAPTER 6:
ECOTOXICOLOGICAL AND RISK ASSESSMENT

6.1 Equilibrium - Partitioning theory

Ecotoxicological risks could be studied by experiments in order to determine the partition coefficients of the nonylphenols between soil, water and sediment. Eight leaching tests were carried out in order to determine the partition coefficients of nonylphenols in water-soil-air media by changing factors as temperature, pH and aeration (see Table 10). In these experiments the same concentration of nonylphenols and internal standard (100 μg / ml in acetone) was added to ~50 g (all of the 8) of samples of unpolluted soil (from Göteborg). The acetone was aerated by evaporation during night (Bjuggren *et al.*, 1999). The samples were thereafter mixed with different amounts of nanopure water (250 and 500ml) in 1L beakers. In two beakers the pH was not neutral, in one the temperature was maintained up to 50°C, two were aerated and one of them had indirect contact with the SPME fibre in order to check the possible emissions to air (see Table 10).

Table 10: Specifications of the 8 leaching tests for determination of partitioning coefficient

Experiment No	Liquid-Solid ratio (L/S)	Time of Exposure (hrs)	pH	Temperature (°C)	Aeration	Special Propose
1	10	24	7	Ambient	No	Higher L/S ratio
2	5	24	7	Ambient	No	Standard sample
3	5	24	4	Ambient	No	Low pH
4	5	24	10	Ambient	No	High pH
5	5	24	7	Ambient	Yes	Aeration effect
6	5	24	7	50°C	No	High temperature
7	5	48	7	Ambient	No	Longer time of exposure
8	5	24	7	Ambient	Yes	Contact with SPME device (emissions to air)

These eight samples were mixed continuously during the whole time of exposure with a special electrical mixer at 60 rpm and with the help of 2 compressed air valves (see Figure 30).

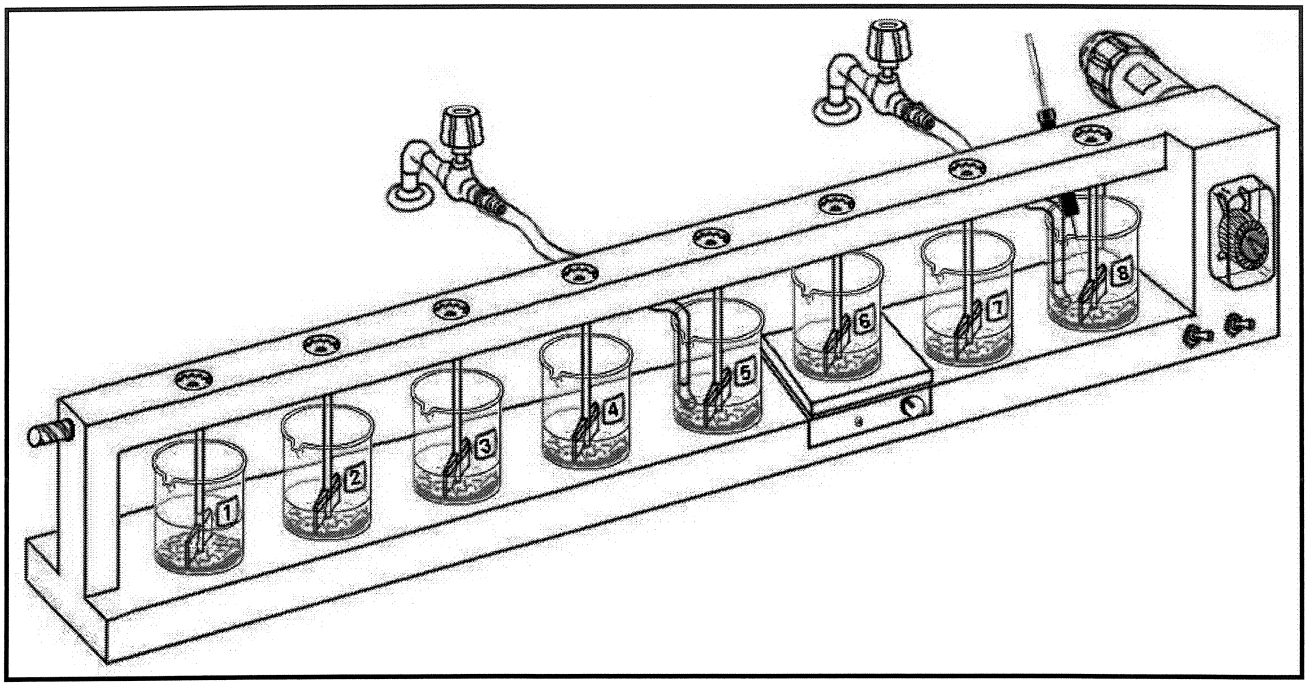


Figure 30: Mixing equipment used to determine partitioning coefficients

After the exposure and mixing time, the samples were submitted to the same filtration process used for the microwave digested samples. A small amount of the liquid (free of solids) was collected (20 ml aprox.) and submitted to the GC analysis after sampling with the SPME device and with the same procedure used for the microwave digested extracts.

Table 11: Concentrations of nonylphenols in the water phase

Experiment No. (SMPE-GC)	A_{NP} / A_{IS}	M_{NP}* in the Water (µg)	[NP] in the Water (µg / ml)	Final volume of Water (ml)
1	0.0081	25 599.8567	51.200	500
2	0.0160	12 917.8106	51.671	250
3	0.0199	12 976.0057	51.904	250
4	0.2082	15 785.7825	63.143	250
5	0.6543	22 442.4018	89.770	~200
6	0.0393	13 265.4888	53.062	~200
7	0.0495	13 417.6913	53.671	250
8**	n.d.	n.d.	n.d.	~200

* Using the same "m" and "b" of the linear model used for the MWD-SPME samples

** No nonylphenols detected in the SPME device put into contact with air

Knowing the amount found in the water extracts and supposing there is no transport to the air, as the experiment 8 proved, the rest of the amount added should be found in the solid soil.

Table 12: Total amount of nonylphenols added, NP in the Soil and its concentration in the soil

Experiment No. (SMPE-GC)	Total Amount added of NP (μg)	M_{NP} in the Soil (μg)	Total weight of Soil used (g)	[NP] in the Soil (μg / mg)
1	50 000	24 400.1433	50.0035	487.968
2	25 000	12 082.1894	50.0994	241.164
3	25 000	12 023.9943	50.0638	240.173
4	25 000	9 214.2175	49.9190	184.583
5	25 000	2 557.5982	49.9577	51.195
6	25 000	11 734.5112	49.9615	234.871
7	25 000	11 582.3087	49.9461	231.896
8*	25 000	n.d.	49.9718	n.d.

* No nonylphenols detected in the SPME device in contact with air over the beaker

The partition coefficient in this case determines the equilibrium of nonylphenols in two different phases: water and soil, but once this compound is present in the aquatic ecosystem it will undergo different processes: volatilization, bioaccumulation, sorption, desorption, sedimentation, resuspension, degradation, transformation, etc. In order to determine the partition coefficient at different conditions it is necessary to use a formula where the equilibrium between these two solvents is dictated by the final concentrations between the two phases:

$$K_{NP} = \frac{[X]_S}{[X]_W}$$

where K_{NP} is the partition coefficient of nonylphenols and $[X]_S$ and $[X]_W$ are the equilibrium concentrations of nonylphenols in the soil and the aqueous phases, respectively. The results clearly indicate that the nonylphenols like better to stay in the non-polar phase as the solid matrix of the soil rather than in the water (see Table 36). It is also obvious that environmental factors could change and increase the transport of nonylphenols to the water. The results show that the highest contribution of this transport is the aeration.

Table 13: Partition coefficients obtained from the leaching test

Experiment No. (SMPE-GC)	Partition Coefficient K_{NP}	Special Propose	Conclusion
1	9.5306	Larger amount of water	Less transport of NP into the water than the standard
2	4.6672	STANDARD	NP prefer to stay in the solid soil phase than in the aqueous
3	3.8034	Acid pH 4	Helps more than neutral pH to get more NP into the water than the standard
4	2.9232	Basic pH 10	Helps more than the acidic and neutral pH to get more NP into the water
5	0.5702	Aeration	Best parameter than any other to get more NP into the water
6	4.4263	Higher Temperature 50°C	Only a little higher transport of NP into the water than the standard
7	4.3206	Longer time of exposure	Only a little higher transport of NP into the water than the high temperature sample
8*	-	Determine transport of NP aqueous solution to the air	No measurable transport of NP in aqueous solution to the air

As a conclusion, the factors that affect the transport of NP most into the water in an equilibrium soil-water are listed below in a decreasing order:

Effect: Aeration > Basic pH > Acid pH > Time of exposure > High temperature > Std > L/S-ratio

Possible Cause: Flooding Chemical Spill Acidification Permanence Global warming Flooding

The future trends and fate of the NP could change from the future scenario presented in Figure 29, where the NP will continue to accumulate into the sediment of the river and reach long distances. From the experiments it is indicated that NP could be emitted from the soil and sediment during periods with flooding or high precipitation, and an aeration process during these periods will emit the NP from the sediment into the water and in this way transport the nonylphenols even longer distances.

6.2 Environmental cycle of organic pollutants

Normally, any persistent organic pollutant emitted into an ecosystem will suffer a slow or fast degradation depending on many factors: Polarity (K_{OW} =partition coefficient of n-octanol-water), available oxygen (DO = dissolved oxygen), pH, biodegradability, toxicity, temperature, concentration, etc. As any other organic substance, nonylphenols could be attractive as food for certain microorganisms whom could start to biodegrade it and transform it into less toxic and more natural found compounds (probably salicylic acid or other carboxylic phenols) which present far low toxicity. This biodegradation could take long time depending on the concentration NP found in the media and on the presence of the favourable microorganisms. Another process that NP could suffer is the slow oxidation process owing to the geochemical properties of the media. In acidic media, nonylphenols suffer a relative high oxidation when a great surface is present and oxygen and high temperature (>30ºC) is present. In alkaline media it would be more stable since it will to some extent be ionized to salts and could have a longer life in the environment. Experiments on the soil have shown that especially NP is rapidly metabolized by aerobic biological mechanism of degradation and that these microorganisms are probably natural widespread in soils. Other microorganisms have been created and isolated to biodegrade nonylphenols more rapidly than the natural species. It is also known that common yeasts as Candida maltosa (Corti *et al.*, 1995) are very useful to biodegrade *p*-nonylphenol at a very high rate (half life of NP [1 mg / L] = 36 h compared to 135 h in normal sewage). The most common mechanism of biodegradation of NP is described below:

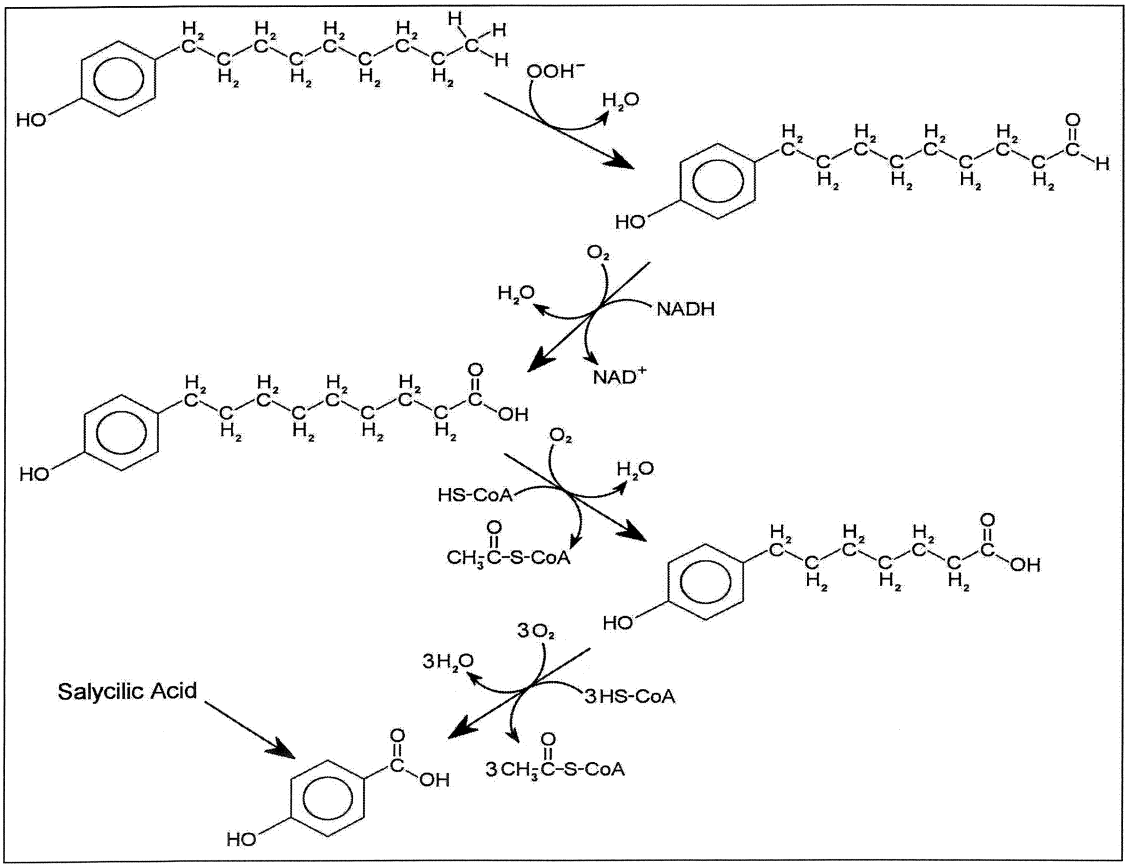


Figure 31: Metabolism of NP in the living cells

6.3 Bioaccumulation factors

The bioaccumulation factors are the relationship between the tendency of one organic compound (normally a pollutant) dissolved in one phase at a low concentration to accumulate in a different phase (living tissue) and reach dangerous or toxic concentrations. Normally, the toxicity is measurable in three levels: acute, chronic and subchronic depending on the toxicity itself of the compound (capacity to make damage to living creatures and metabolisms) and the concentration present in the media (Soil, Sediment, Water, Air). Sometimes the concentration reaches very high levels but with no appreciable major damage until the death of the organism. But normally, there is a common limit of concentration where the largest damage and effect occur, sometimes with no visual or measurable action before this level. The bioconcentration factor (BCF) describes the ratio of the concentration of a pollutant in a living organism and the same concentration in the water after a certain period of time. Usually the laboratories determine this value making experiments with some invertebrate animals as mollusks, insects, bacteria, etc or fishes.

The experimental value for nonylphenol is:

BCF in marine mussel (Mytilus edulis): 10*
*With saturated NP aqueous solution @ NTP = 0.953 g / 100ml

To get an idea of comparison, Table 29 presents a comparison of the BCF of NP with a high persistent pollutant as DDT and a very low factor such as for Benzoic acid.

Table 14: Bioconcentration factor (BCF) of different environmental pollutants

Compound	BCF
Hexachlorobenzene	275
DDT	120
p-Nonylphenol	10
Phenol	5.5
Benzoic Acid	1.25

This value indicates only the capacity to be accumulated in a certain organism, and the value should be studied together with the toxicity reached at this concentration because sometimes a compound with a higher value of BCF could make less damage to the living tissue than nonylphenols themselves. The toxicity of nonylphenols contact, exposure and possible routes of distribution and bioaccumulated concentrations are studied in the next subchapter.

6.4 Biodiversity and human health

As a base for estimation of the toxicity of nonylphenols its physical and chemical properties from its MSDS (Material Safety Data Sheet) (Akzo Nobel et al,. 2000) (see Appendix) could be studied. Other effects have also to be also considered for a complete evaluation. Nonylphenols as alkylphenol compounds, are proven to be environmental estrogens, substances with the ability to act like estrogen hormones in living organisms. Humans can carry some of this kind of chemicals in fat and tissue and could deliver them to the children during pregnancy and breastfeeding. A literature report indicates a first fast elimination of nonylphenols in the liver after oral ingestion and an accumulation process could only happen if these paths are saturated (Certa *et al.*, 1994). Concerning the biodiversity health, experiments carried out (Soto *et al.*, 1995) showed that the alkylphenol compounds are concentrated by organisms such as fish and birds, leading to an accumulation in their organs between ten to several thousand times greater than in the surrounding environment. One example of these studied animals is the rainbow trout (Oncorhynchus myskiss), where analysis have concluded that estrogenic (estrogen-mimicking) effects are present at tissue concentration of 220 µg NP or even lower concentrations. Besides fishes and birds, aquatic flora have also been shown accumulation of NP up to levels of 10 000 greater than in the surrounding environment, causing toxic effect in the germination and growth of different species.

Table 15: Comparison of the value obtained in this work with the maximum projected threshold concentration of some soil pollutants according to Dutch Authorities

Type of Compound	Threshold (mg / kg)	Pollutant Common Sources	Occurrence
POPs, sum	8	Creosote Coal Tar Oil Derivates	Wood Treatment Lubrication Industry Surfactant Industry
POP = NP*	In this study: **23 (average)**		
PCBs, sum	0.2	Transformer Oil Hydraulic Oil Insecticides	Electrical Industry Metal Industry Healthcare Industry
Heavy Metals	5 – 5000 (depending on the metal)	Coal Tar Wood Various	Electrical Industry Wood Treatment Metal Industry

*Result obtained in this work

The value reported for NP in Table 15 is beyond one of the European suggested threshold limit for the total concentration of POPs that should be found in soil environments. Extreme cautions and measurements are recommended for a quick remediation to avoid more dangerous and specific changes in living organism such as hormonal interaction.

CHAPTER 7:
RESULTS MANAGEMENT AND PROPOSED METHODOLOGY

7.1 Physical and chemical methods

Today, in a global market, there are many methods available for the physical remediation of soils, river water and sediments. These proposals includes techniques from the removal with a extraction, oxidation onsite and destruction of the pollutant with chemicals.

The extraction remediation process uses non-toxic and environmental friendly solvents (even water) to remove at maximum the pollutants found in a certain amount of soil and sometime in sediment. This process is based on the same physical principles described before for the Soxhlet extraction. The methodology is very easy: a special equipment is brought to the site to carry and put the soil or sediment into a big tank. Then solvent is added to the tank trying to get into contact with as large surface as possible (see Figure 32). The mixture is stirred for a certain time and filtered and the solid is collected as a clean soil or sediment. Sometimes the whole process is repeated many times to get as high efficiency as possible. The solid is returned again into its original space. The advantages of this method are the cheap cost and easiness to implement. The drawbacks are the difficult selection of an adequate solvent to a specific pollutant, the low efficiency obtained in the extraction and the amount of solid should be as small as possible to be economically feasible (Oliviera *et al.*, 1999)

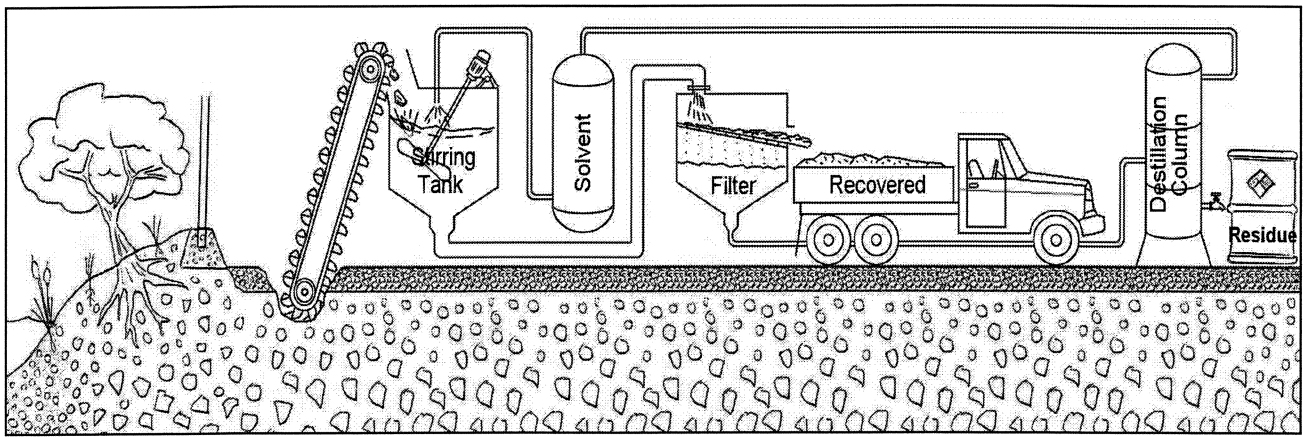


Figure 32: Description of the extraction physical process for cleaning of contaminated soil

The oxidation process (known also as the aeration process) is based on the principle of reaction between oxygen of the air and the pollutant in the solid surface of the soil or sediment put in a bed with holes of certain size, allowing only the pass of the air but not of the solid particles (see Figure 33). This process could take more time than the extraction process but with no production of a residue concentrate and use of any solvent. The main advantages of this process is the use of only atmospheric air with forced draft bed fan, low operational cost and no need to extreme control. The main disadvantages are the large time the process could take to oxidize the pollutant (specially if there is some chlorinated compound), sometimes the efficiency is very low, and the further degradation of the natural resources in the soil or sediment. There is also a risk for volatilization of the organic pollutants causing air pollution problems. Aeration is the method used nowadays for cleaning the soil at the Akzo Nobel Mölndal plant area.

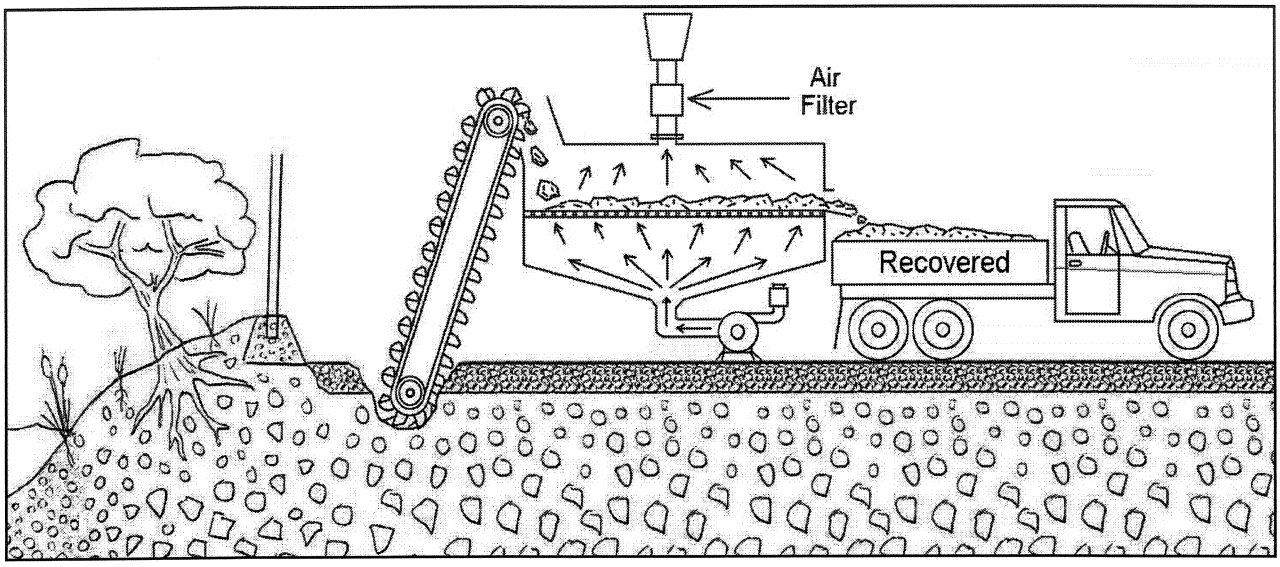


Figure 33: Description of the oxidation or aeration physical process for cleaning of contaminated soil

The last physical methodology for remediation is the least used because it implies the use of more chemicals and sometimes they could be also hazardous for the environment. The main principle is a chemical attack to the pollutant with liquid or liquid solutions of substances able to destroy (normally oxidizing), and a light extraction of the residue and products of this reaction. The chemicals used are: chlorine, chlorine oxides solutions, diluted chlorates and sulfuric acid, diluted hydrogen peroxide, aqueous solutions of ozone, etc. The main advantages for this process are: high destruction of the most of the pollutants, low residence time in all equipments, and fast installation of the soil treated. The main disadvantages are: the possible addition to the soil or sediment with even more dangerous substances, expensive cost if a large amount of soil should be treated, and possible damage to the natural structure of the solid matrix.

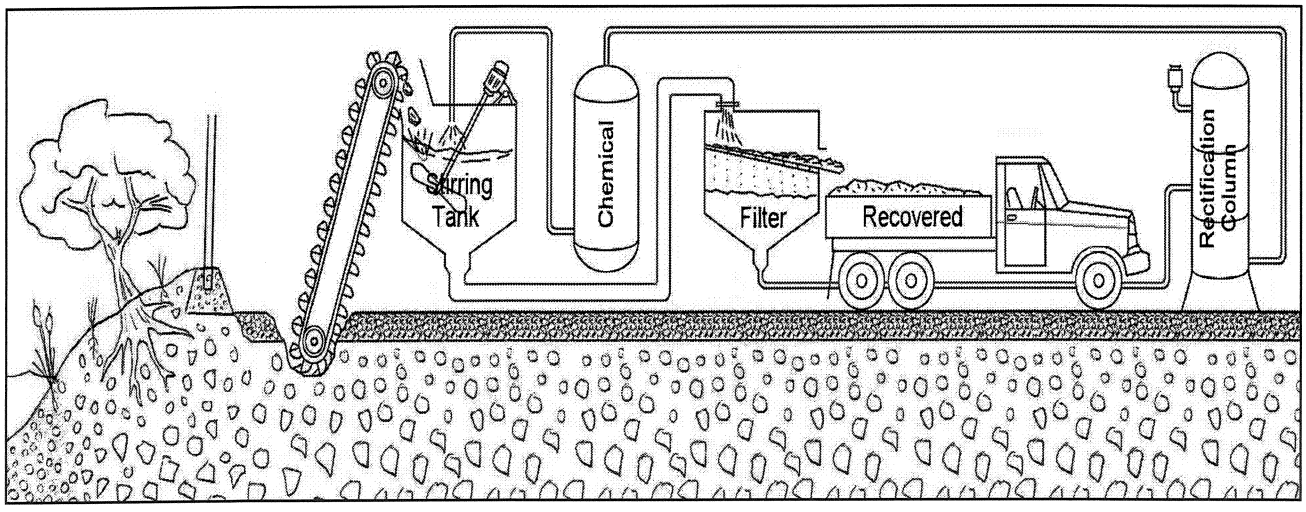


Figure 34: Brief description of the Chemical Process

7.2 Biochemical methods

Many biochemical methods have been reported in the literature to remediate the problem of soil and sediment pollution. The oldest ones use the principle of a possible degradation and metabolization of pollutants with normal and less specific plants (Phytoremediation). Phytoremediation is the use of plants and trees to clean up contaminated soil, sediment and water. Using plants to remediate contaminated medias is an aesthetically pleasing, solar energy driven, passive technique suitable for a variety of pollutants and conditions. The advantages of this process are: the potentially marketable by-products, low impact and passive, safe to implement, and natural plants can aid in site restoration (NCER *et al.*, 2000). The main disadvantages are: the possible failure of the plants to metabolize a very specific and low degradable pollutant, long period of time to gain reliable results, and possible production of undesirable by-products (more chlorinated compounds).

There are other kinds of biochemical methods cited in the literature which describes a more potential use of some microorganisms. These methods are based in specific or created microorganisms which are able to find a major route in their metabolism the pollutants. Some of these microorganisms are very common and naturally found. Special care must be taken of created organism not to make further damage or lead to an incontrollable growth. The use of microorganisms to destroy the pollutant in soil or sediment is very easy to implement. They just have to be grown in a lab in cepas and then just sought into the media in situ. Nevertheless, there are some methods which collect all the involved soil or sediment and treat them in a separate reactors and plants, but of course they are more expensive and present a solution for a very polluted soil or medias polluted with a highly toxic pollutants. Two groups of methods exist according to type of metabolism: aerobic and anaerobic degradation. Microorganisms consuming oxygen for their metabolism belong to the first group. They practically "burn the compounds with oxygen" to gain energy. The second group works in very poor concentration of oxygen and even in a total lack of this gas, and they gain the energy transforming the compounds into a smaller molecules only. The suggestions found in the literature describe more aerobic treatments for pollutants than anaerobic (20:1) and many authors and investigators consider them more suitable to do the job (Plomley *et al.*, 1999)

The recognized microorganisms, which present a real and effective activity on nonylphenols, have mainly aerobic metabolism as fungi and bacteria. As mentioned before, *Candida maltosa* presents a high degradation rate and its efficiency has been proved even when it is implemented in sewage plants and in situ. Another useful microorganism is a similar yeast, *Candida aquatexterois* (Vallini *et al.*, 2001), which is a rarer species than the *maltosa*, but could present a more stable growth and control in the metabolism of NP. In the literature a more detailed description of the process, mechanisms and by-products of this digestion is found. Some of the final compounds are: 4-hydroxy-benzoic acid, *cis* and *trans*-4-hydroxy-cinnamic acids, 4-hydroxy-acetophenone and 3-(4-hydroxyphenyl)-propionic acid. These compounds present a 1000 times less acute toxicity to the environment than NP itself.

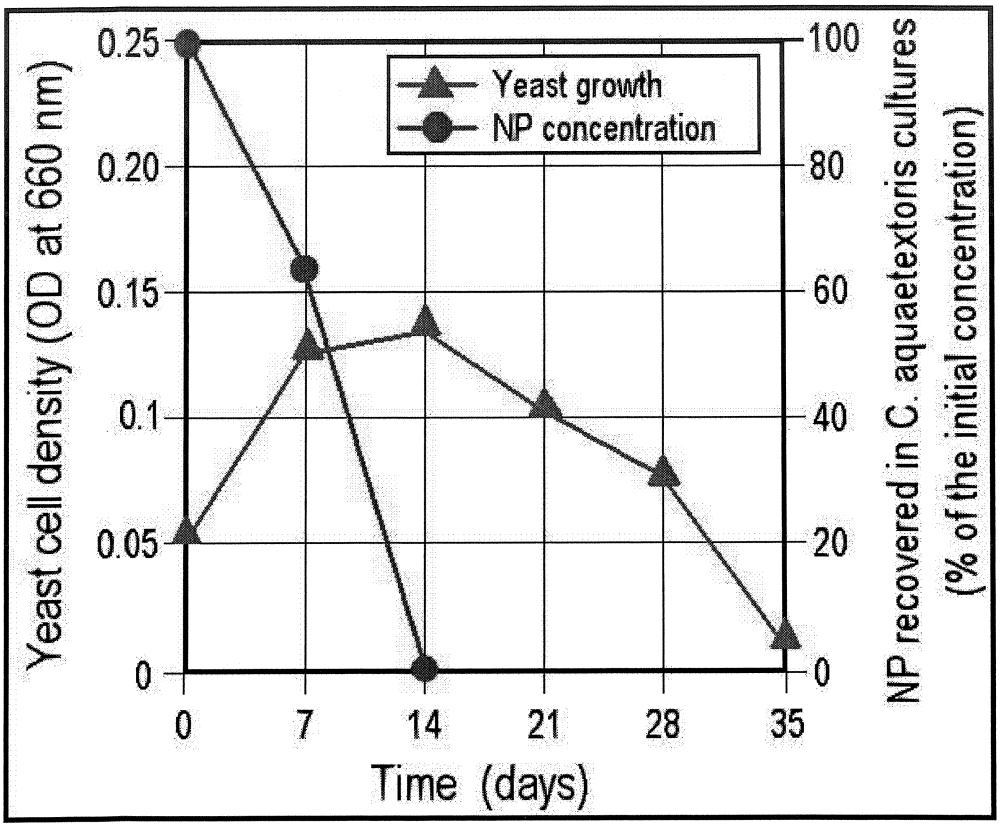


Figure 35: Dynamics of NP utilization by Candida aquaetextoris during growth in yeast base (Vallini *et al.*, 2001)

Except the species mentioned, there are many others recommended by companies and governmental authorities to present a reliable and environmental friendly solutions for remediation of media polluted with nonylphenols. A suggested further study of these possibilities is a warranty to solve the present problem in the Mölndal river sediment, soil and water.

7.3 Policy of authorities

All over the world different governmental policies exist for a remediation of polluted areas, e.g. economical punishments, but the laws change from country to country. A more detailed study of the local legislation is a good beginning before input of any possible repercussion. Historically, potential surfactant contamination (NP included) followed the shift from the use of soap-based detergents to more complex surfactants. Restrictions on the use of these substances have arisen since the discovery in 1984 that some of their breakdown products are more toxic to aquatic life than the surfactants themselves. These problems lead to bans and restrictions on the use of phenolic surfactants in household and industrial cleaning applications in Europe. The environmental agencies policy of some European countries (Germany, Switzerland, Sweden, Danmark, France) is now concerned in the side effects these products, such as

more solubilization of other fatty-soluble pollutants as DDT and trichlorobenzene in water with the help of the surface-active properties of these compounds.

A study in the Netherlands of the risk to the aquatic environment from a range of surfactants established a specific parameter called PEC (predicted environmental concentration) for a distance of 1000m below the sewage outfall. This parameter is related to another called PNEC (predicted no effect concentration). The study revealed a ratio of PEC/PNEC of 2.5 for surfactants gathered from several locations and supplied by Dutch industries. This value suggests that a high risk to the aquatic environment is expected. Another study carried out in Germany show a half life of 25 days can be expected for nonylphenols (4.7mg/kg) in polluted soil. (Litz *et al.*, 1992)

After too many studies, the European Commission for the Environment is very concerned trying to develop a reasonable policy to fulfill two main objectives: Prohibition law on the synthesis of the most dangerous surfactants materials (NP included) and implementation of alternatives to industries to remediate already polluted areas and ecosystems. One example of a strict control in the subject was carried out by the British Environmental Agency, where many companies were economical subjected to cover the costs for a remediation of large (1ha) industrial areas. The solutions presented by these authorities included sewage sludge disposal, soil bioremediation, landfill and incineration methods. Pressure from environmentalists and rising costs are gradually making landfill and incineration less attractive. This combined with increasing amounts of sludge produced, is increasing the pressure on agricultural application. It appears that surfactant application to aerobic soils is quite safe due the rapid biodegradation rates. However, the temptation to dispose of sludge on non-agricultural soils should be carefully investigated. Soils that are anaerobic may not be appropriate sites for amendment. Such soils may accumulate surfactants since biodegradation is retarded and may result in surfactant global contamination of the environment.

A conscious study of the governmental policies applied for Sweden is recommended to follow the possible routes and solutions to this delicate problem.

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APPENDIX

Tables of the results from the weighting of Sediment:

Samples before the Soxhlet extraction:

1. Weight of : vase + empty paper sampler
2. Weight of : vase + paper sampler + sample
3. Weight of : pure sample (20 g aprox.)

Samples before determination of water, organic and inorganic content:

1. Weight of : empty crisol
2. Weight of : crisol + sample
3. Weight of : pure sample (5 g aprox.)

Samples after determination of water, organic and inorganic content:

1. Total Weight of the sample
2. Water content
3. Dry Weight (Organic + Inorganic)
4. Weight of Organic Matter
5. Weight of Inorganic Matter
6. % Water
7. % Dry Solid (Organic + Inorganic)
8. % Organic Matter
9. % Inorganic Matter

Table 1: Total weight of sediment samples before Soxhlet extraction

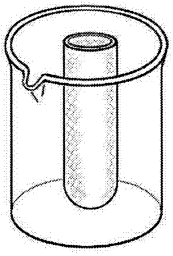
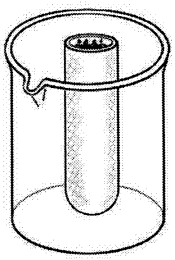
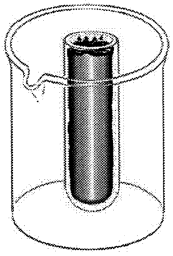
Sediment Sample No.			
1	76.862 g	98.797 g	21.935 g
2	67.372 g	87.400 g	20.028 g
3	77.180 g	97.421 g	20.241 g
4	73.823 g	94.132 g	20.309 g
5	64.800 g	94.309 g	20.740 g
6	73.792 g	94.309 g	20.517 g
7	65.305 g	85.354 g	20.049 g
8	76.002 g	96.063 g	20.061 g
9	76.412 g	96.472 g	20.060 g
10	64.825 g	84.490 g	19.665 g
11	75.256 g	95.450 g	20.194 g
12	74.446 g	94.490 g	20.044 g

Table 2: Total weight of sediment samples before determination of water, organic and inorganic content

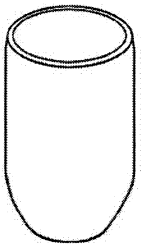
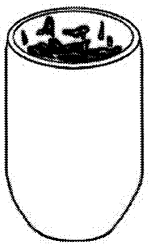
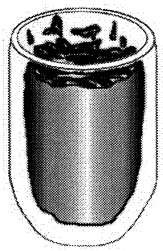
Sediment Sample No.			
1	25.81080 g	30.06420 g	4.25340 g
2	27.78871 g	32.60311 g	4.81440 g
3	27.47962 g	32.61042 g	5.13080 g
4	27.75310 g	32.71411 g	4.96101 g
5	27.76155 g	32.92785 g	5.16630 g
6	25.75530 g	30.75031 g	4.99501 g
7	25.69103 g	30.68696 g	4.99593 g
8	25.86788 g	31.37899 g	5.51111 g
9	27.39780 g	32.45705 g	5.05925 g
10	14.16520 g	19.21860 g	5.05340 g
11	17.12040 g	22.14309 g	5.02269 g
12	16.51743 g	21.59202 g	5.07459 g

Table 3: Total weight, water content, dry weight, organic and inorganic matter of sediment samples

Sediment Sample No.	Total Weight	Water Content	Dry Weight (Organic & Inorganic)	Organic Matter	Inorganic Matter
1	4.25340 g	2.20691 g	2.04649 g	0.07664 g	1.97026 g
2	4.81440 g	2.88735 g	1.92705 g	0.09455 g	1.83250 g
3	5.13080 g	3.03459 g	2.09621 g	0.11697 g	1.97924 g
4	4.96101 g	1.76200 g	3.19901 g	0.18179 g	3.01722 g
5	5.16630 g	2.57975 g	2.58655 g	0.12812 g	2.46053 g
6	4.99501 g	3.50910 g	2.09621 g	0.23262 g	1.86359 g
7	4.99593 g	2.98477 g	2.01116 g	0.08795 g	1.93488 g
8	5.51111 g	3.49619 g	2.01492 g	0.20360 g	1.81132 g
9	5.05925 g	3.03642 g	2.02283 g	0.16801 g	1.86359 g
10	5.05340 g	1.05050 g	4.00290 g	0.19556 g	3.80734 g
11	5.02269 g	1.48521 g	3.53745 g	0.13421 g	3.40324 g
12	5.07459 g	3.38253 g	1.69206 g	0.20339 g	1.48867 g
13	1.8797 g	0.9889 g	0.8907 g	0.0630 g	0.8276 g
14	1.8937 g	0.9913 g	0.9023 g	0.0700 g	0.8322 g
15	1.5182 g	0.5752 g	0.9429 g	0.0367 g	0.9061 g
16	0.8762 g	0.3836 g	0.4925 g	0.0196 g	0.4728 g
17	3.3893 g	1.2245 g	2.1647 g	0.0690 g	2.0956 g
18	2.0268 g	1.2576 g	0.7691 g	0.1013 g	0.6677 g
19	3.0193 g	0.9184 g	2.1008 g	0.0338 g	2.0669 g

Table 4: Percentage of total weight, water content, dry weight, organic and inorganic matter of sediment samples

Sediment Sample No.	% Water	% Dry Solid (Organic & Inorganic)	% Organic Matter	% Inorganic Matter
1	51.8858	48.1142	1.8101	46.3220
2	59.9733	40.0267	1.9638	38.0628
3	59.1446	40.8554	2.2797	38.5756
4	35.5168	64.4832	3.6643	60.8186
5	49.9342	50.0658	2.4799	47.6265
6	70.2522	29.7478	4.6570	37.3090
7	59.7441	40.2559	1.7604	38.4955
8	63.4390	36.5610	3.6943	32.8667
9	60.0172	39.9828	3.3208	36.8353
10	20.7880	79.2120	3.8698	75.3421
11	29.5702	70.4298	2.6720	67.7531
12	66.6563	33.3437	4.0080	29.3357
13	52.61	47.39	7.08	92.92
14	52.35	47.65	7.76	92.24
15	37.89	62.11	3.90	96.10
16	43.79	56.21	3.98	96.02
17	36.13	63.87	3.19	96.81
18	62.05	37.95	13.18	86.82
19	30.42	69.58	1.61	98.39

Tables of the results from the weighting of Soil:

Samples before the Soxhlet extraction:

1. Weight of : vase + empty paper sampler
2. Weight of : vase + paper sampler + sample
3. Weight of : pure sample (20 g aprox.)

Samples before determination of water, organic and inorganic content:

1. Weight of : empty crisol
2. Weight of : crisol + sample
3. Weight of : pure sample (5 g aprox.)

Samples after determination of water, organic and inorganic content:

1. Total Weight of the sample
2. Water content
3. Dry Weight (Organic + Inorganic)
4. Weight of Organic Matter
5. Weight of Inorganic Matter
6. % Water
7. % Dry Solid (Organic + Inorganic)
8. % Organic Matter
9. % Inorganic Matter

Table 5: Total weight of soil samples before Soxhlet extraction

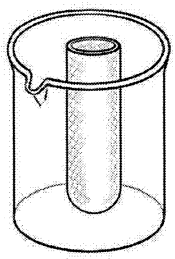
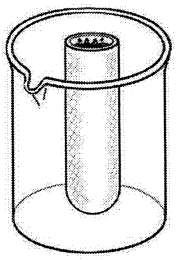
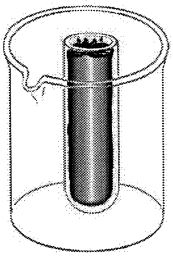
Soil Sample No.			
1	140.537 g	160.751 g	20.214 g
2	140.823 g	160.830 g	20.007 g
3	140.802 g	160.943 g	20.141 g
4	140.733 g	161.035 g	20.302 g
5	141.213 g	161.408 g	20.195 g
6	139.473 g	159.794 g	20.321 g
7	74.647 g	94.684 g	20.037 g
8	74.867 g	94.861 g	19.994 g
9	140.307 g	160.753 g	20.446 g
10	139.319 g	159.765 g	20.444 g

Table 6: Total weight of soil samples before determination of water, organic and inorganic content

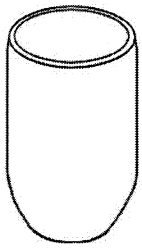
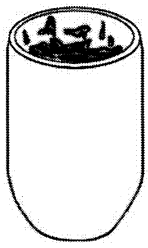
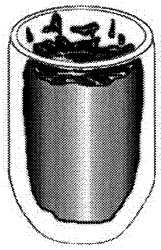
Soil Sample No.			
1	27.39675 g	32.32625 g	4.92950 g
2	25.86769 g	30.93520 g	5.06751 g
3	25.69063 g	30.76955 g	5.07887 g
4	26.31273 g	31.38842 g	5.07569 g
5	26.85311 g	31.90027 g	5.04716 g
6	14.16544 g	19.23399 g	5.06855 g
7	25.75413 g	30.72092 g	4.96679 g
8	27.76165 g	32.83521 g	5.07356 g
9	17.12045 g	22.63940 g	5.51895 g
10	16.51645 g	21.76785 g	5.25140 g

Table 7: Total weight, water content, dry weight, organic and inorganic matter of soil samples

Soil Sample No.	Total Weight	Water Content	Dry Weight (Organic & Inorganic)	Organic Matter	Inorganic Matter
1	4.92950 g	1.21564 g	3.71386 g	0.99835 g	2.71551 g
2	5.06751 g	1.15671 g	3.91080 g	0.86753 g	3.04327 g
3	5.07887 g	1.00894 g	4.06993 g	1.28730 g	2.78263 g
4	5.07569 g	0.92275 g	4.15294 g	1.00076 g	3.15218 g
5	5.04716 g	1.11952 g	3.92764 g	1.17992 g	2.74772 g
6	5.06855 g	0.08575 g	4.98280 g	1.08441 g	3.89839 g
7	4.96679 g	1.80047 g	3.16632 g	1.13440 g	2.03192 g
8	5.07356 g	1.62223 g	3.45133 g	0.77029 g	2.68104 g
9	5.51895 g	1.04997 g	4.46898 g	0.90202 g	3.56696 g
10	5.25140 g	0.99978 g	4.25162 g	1.33701 g	2.91461 g

Table 8: Percentage of total weight, water content, dry weight, organic and inorganic matter of soil samples

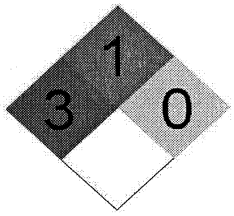
Soil Sample No.	% Water	% Dry Solid (Organic & Inorganic)	% Organic Matter	% Inorganic Matter
1	24.6605	75.3394	20.2525	55.0869
2	22.8260	77.1739	17.1194	60.0545
3	19.8654	80.1345	25.6431	54.4914
4	18.1797	81.8202	19.7167	62.1035
5	22.1811	77.8188	23.3778	54.4410
6	1.6918	98.3081	21.3948	76.9133
7	36.2501	63.7498	22.8397	40.9101
8	31.9736	68.0263	15.1824	52.8439
9	19.0248	80.9751	16.3440	64.6311
10	19.0383	80.9616	25.4600	55.5016

Material Safety Data Sheet of Nonylphenols:

NONYL PHENOL (mixed isomers)
CAS No.: 25154-52-3

Synonyms: 2,6-dimethyl-4-heptyl phenol
Molecular Formula: $C_{15}H_{24}O$
Molecular Weight: 220.39
Physical Aspect: Clear, straw colored, viscous liquid with a slightly phenolic odor
Boiling Point: 293-297°C
Pouring Point: 2°C
Viscosity: 1690 cP @ 25°C
Vapour density: 7.59 (air = 1)
Flash Point: 140.5°C
Density: 0.949 @ 20°C
Solubility: In water = 0.953 g / 100ml. More soluble in aqueous NaOH, Miscible in benzene, chlorinated solvents, aniline, heptane, aliphatic alkanes, ethylene glycol.
General Safety Profile: Moderately toxic by ingestion and skin contact. A severe skin and eye irritant. Combustible when exposed to heat or flame. When heated to decomposition it emits acrid smoke and irritating fumes.
H.M.I.S. Ratings (HMIS = Hazardous Material Identification System)

HEALTH: 3
FLAMMABILITY: 1
REACTIVITY: 0
SPECIAL RISKS: None
Carcinogenicity: Not listed by ACGIH, IARC, NIOSH or OSHA as carcinogen agent



The next step is to analyze the reported toxicities found in the literature. For toxic doses three values were found:

LD_{50} : 1620 mg/kg (oral rat) UCDS (Union Carbide Data Sheet)
LD_{50} : 2140 mg/kg (skin rabbit) AIHAAP (American Industrial Hygiene Association Journal)
LD_{50} : 1231 mg/kg (oral mouse) NTIS (National Technical Information Service)
(LD_{50} = Lethal Dose to reach the 50% of casualties in the tested animals)
For Primary Irritation Effects three values were found:

SEV : 10 mg/24 h (skin rabbit) AMIHBC (Archives of Industrial Hygiene Occupational Medicine)
MOD: 500 mg (skin rabbit) NTIS (National Technical Information Service)
SEV : 50 µg (eye rabbit) AMIHBC (Archives of Industrial Hygiene Occupational Medicine)
Ecotoxicological Data:
BCF in marine mussel (Mytilus edulis): 10 (saturated NP aqueous solution @ NTP = 0.953g/100ml)
Biohazard category: Harmful to any specie (Hormone disrupter)

Concentration and areas obtained in the chromatographic analysis

Table 9: Concentration and total area of the STANDARD I Sample (in heptane)

Compounds	Concentration (in Heptane)	Total Area
Internal Standard (1-fluoro-biphenyl)	25 µg / ml	800 700
Nonylphenols	25 µg / ml	807 170

Table 10: Concentration and total area of the STANDARD II Sample (in heptane)

Compounds	Concentration (in Heptane)	Total Area
Internal Standard (1-fluoro-biphenyl)	25 µg / ml	580 561
Nonylphenols	50 µg / ml	1 054 910

Table 11: Concentration and Total Area of the STANDARD III Sample (in heptane)

Compounds	Concentration (in Heptane)	Total Area
Internal Standard (1-fluoro-biphenyl)	25 µg / ml	655 729
Nonylphenols	100 µg / ml	2 687 665

Table 12: Sediment samples (SOXHLET) total areas obtained from the chromatograms

Sediment Sample (SOXHLET) No.	Total Area of Internal Standard	Total Area of Nonylphenols	Ratio between A_{NP} / A_{IS}
1	183 806	639 003	3.4765
2	338 967	82 191	0.2424
3	1 019 037	103 800	0.1018
4	This was only	submitted to the	GC-MS analysis
5	947 081	591 963	0.6250
6	1 549 011	4 429 399	2.8595
7	12 411 341	1 231 535	0.0992
8	714 654	267 222	0.3739
9	479 599	120 618	0.2514
10	1 854 005	673 541	0.3632
11	698 372	7 091 418	10.1542
12	857 069	101 544	0.1184

Table 13: Soil samples (SOXHLET) total areas obtained from the chromatograms

Soil Sample (SOXHLET) No.	Total Area of Internal Standard	Total Area of Nonylphenols	Ratio between A_{NP} / A_{IS}
1	318 206	2 306 943	7.2498
2	2 912 408	3 592 699	1.2335
3	1 213 066	1 545 138	1.2737
4	1 001 743	510 874	0.5099
5	1 436 266	3 672 077	2.5566
6	4 030 680	1 036 785	0.2572
7	2 016 131	10 922 027	5.4173
8	2 043 101	872 399	0.4269
9	2 138 436	604 373	0.2826
10	1 690 549	465 318	0.2752

Table 14: Concentration and total area of the STANDARD I sample in water

Compounds	Concentration (in Water)	Total Area
Internal Standard (1-fluoro-biphenyl)	25 µg / ml	652 560
Nonylphenols	25 µg / ml	650 799

Table 15: Concentration and total area of the STANDARD II sample in water

Compounds	Concentration (in Water)	Total Area
Internal Standard (1-fluoro-biphenyl)	25 µg / ml	630 064
Nonylphenols	50 µg / ml	1 359 585

Table 16: Concentration and total area of the STANDARD III sample in water

Compounds	Concentration (in Water)	Total Area
Internal Standard (1-fluoro-biphenyl)	25 µg / ml	505 607
Nonylphenols	100 µg / ml	2 198 415

Table 17: Sediment samples (MICROWAVE DIGESTION - SPME) total areas obtained from the chromatograms

Sediment Sample (MWD - SPME) No.	Total Area of Internal Standard	Total Area of Nonylphenols	Ratio between A_{NP} / A_{IS}
1	197 247	634 050	3.2145
2	418 551	99 824	0.2385
3	1 164 773	117 758	0.1011
4	781 054	202 895	0.2597
5	1 650 762	965 035	0.5846
6	2 935 685	7 733 917	2.6344
7	10 600 526	971 008	0.0916
8	975 502	353 814	0.3627
9	465 217	112 629	0.2421
10	1 054 372	361 122	0.3425
11	1 176 198	10 741 746	9.1326
12	1 840 127	196 525	0.1068
13	801 569	412 707	0.5148
14	1 007 813	212 547	0.2109
15	1 157 280	383 869	0.3317
16	326 950	1 013 512	3.0999
17	733 974	141 583	0.1929
18	589 661	653 580	1.1084
19	697 227	282 920	0.4057

Table 18: Soil samples (MICROWAVE DIGESTION - SPME) total areas obtained from the chromatograms

Soil Sample (MWD - SPME) No.	Total Area of Internal Standard	Total Area of Nonylphenols	Ratio between A_{NP} / A_{IS}
1	632 338	4 316 023	6.8255
2	9 159 523	10 446 604	1.1403
3	2 604 452	2 964 388	1.1382
4	1 998 477	943 281	0.4720
5	1 369 623	3 294 902	2.4057
6	8 119 804	1 884 606	0.2321
7	3 760 890	19 660 432	5.2276
8	1 409 208	541 417	0.3842
9	3 354 564	923 176	0.2752
10	3 045 439	782 068	0.2568

Table 19: Water samples (SPME) total areas obtained from the chromatograms (No IS was added to these samples)

Water Sample (SPME) No.	Total Area of Nonylphenols
1	Not Detected
2	Not Detected
3	Not Detected
4	Not Detected
5	Not Detected
6	Not Detected
7	Not Detected
8	Not Detected
9	Not Detected

Table 20: A_{NP} / A_{IS} relationship and total weight of nonylphenols (M_{NP}) in sediment (Soxhlet) solvents extracts

Sediment Sample (SOXHLET) No.	A_{NP} / A_{IS}	M_{NP} (µg)
1	3.4765	137.8114
2	0.2424	33.1683
3	0.1018	28.6190
4*	6.9324	249.6311
5	0.625	45.5477
6	2.8595	117.8476
7	0.0992	28.5349
8	0.3739	37.4231
9	0.2514	33.4595
10	0.3632	37.0769
11	10.1542	353.8762
12	0.1184	29.1564

*This sample was submitted to GC-MS analysis

Table 21: A_{NP} / A_{IS} relationship and total weight of nonylphenols (M_{NP}) in soil (Soxhlet) solvent extracts

Soil Sample (SOXHLET) No.	A_{NP} / A_{IS}	M_{NP} (µg)
1	7.2498	259.9009
2	1.2335	65.2365
3	1.2737	66.5372
4	0.5099	41.8235
5	2.5566	108.0469
6	0.2572	33.6471
7	5.4173	200.6082
8	0.4269	39.1380
9	0.2826	34.4690
10	0.2752	34.2295

Table 22: A_{NP} / A_{IS} relationship and total weight of nonylphenols (M_{NP}) in sediment (MWD-SPME) water extracts

Sediment Sample (MWD - SPME) No.	A_{NP} / A_{IS}	M_{NP} (µg)
1	3.2145	121.2904
2	0.2385	32.4758
3	0.1011	28.3753
4	0.2597	33.1085
5	0.5846	42.8047
6	2.6344	103.9781
7	0.0916	28.0917
8	0.3627	36.1824
9	0.2421	32.5832
10	0.3425	35.5795
11	9.1326	297.9790
12	0.1068	28.5454
13	0.5148	40.7216
14	0.2109	31.6521
15	0.3317	35.2572
16	3.0999	117.8703
17	0.1929	31.1149
18	1.1084	58.4367
19	0.4057	37.4656

Table 23: A_{NP} / A_{IS} relationship and total weight of nonylphenols (M_{NP}) in soil (MWD-SPME) water extracts

Soil Sample (MWD - SPME) No.	A_{NP} / A_{IS}	M_{NP} (μg)
1	6.8255	246.1722
2	1.1403	62.2209
3	1.1382	62.1529
4	0.472	40.5972
5	2.4057	103.1644
6	0.2321	32.8350
7	5.2276	194.4703
8	0.3842	37.7564
9	0.2752	34.2295
10	0.2568	33.6342

Calibration techniques

The method with internal standard is the most used method for quantification in chromatography, since it requires individual injections to yield reliable results. For the calculation of the concentration of the analytes found by the chromatographic analysis, the next formula is needed (Reece *et al.*, 1989):

Amount of nonylphenols (μg) per amount of sediment-soil-water (g) = $Q = \left[\frac{\mu g}{g}\right]$

$$Q = \frac{\dfrac{\text{Peak area of NP in the sample solution}}{\text{Peak area of IS in the sample solution}} \text{ X Weight of the IS added (μg) X \% Purity of Standard}}{\dfrac{\text{Peak area of NP in the standard solution}}{\text{Peak area of IS in the standard solution}} \text{ X Weight of the sample (g)}}$$

The relation between the nonylphenols and IS areas in the standard solution should be normalized and coupled to mathematical model. The standard samples were analyzed to estimate the concentrations of the analytes. A graph for the calibration of the relation was carried out. Three calibrates in heptane for Soxhlet and three in water for MWD. These graphs showed a linear tendency behavior when plotting the Peak Area of IS in the Standard solution in the x-axis and the peak area of NP in the Standard solution in the y-axis, and then we coupled a mathematical model of $y = mx + b$.

The next two graphs show the relationship between the area of nonylphenols and internal standard:

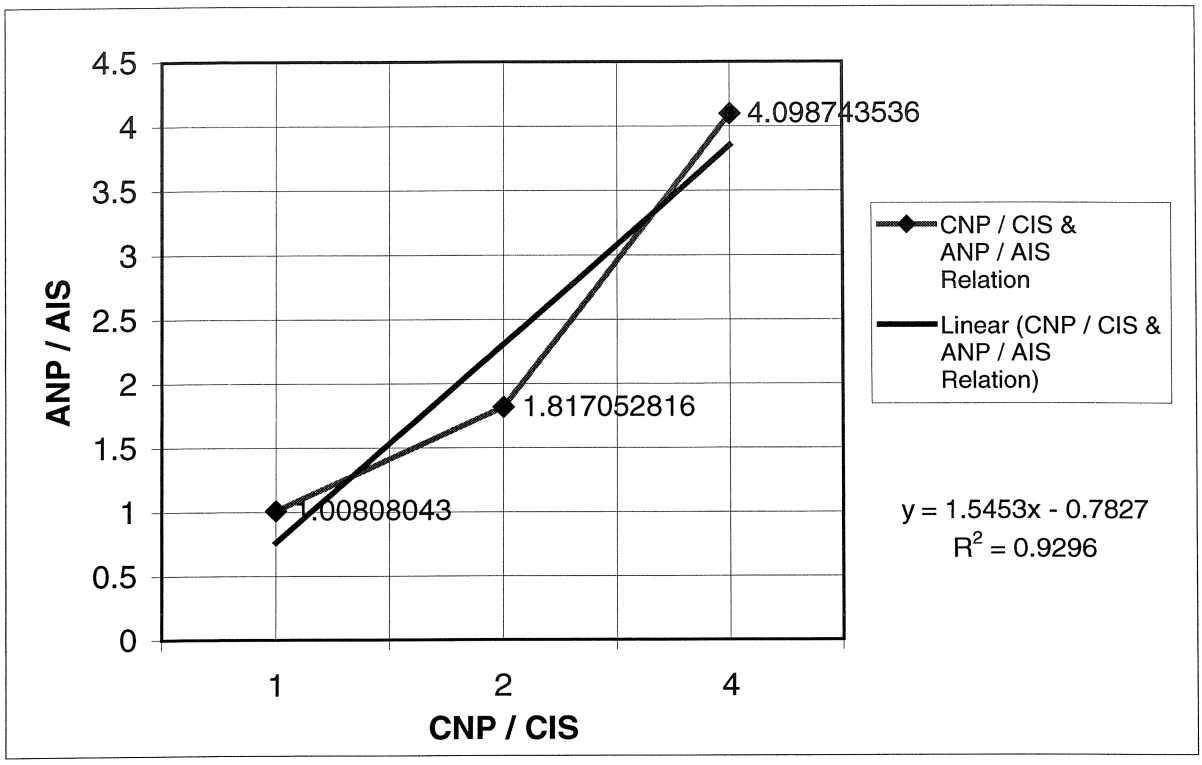


Figure 1: Calibration curve for nonylphenols in solvent extracts

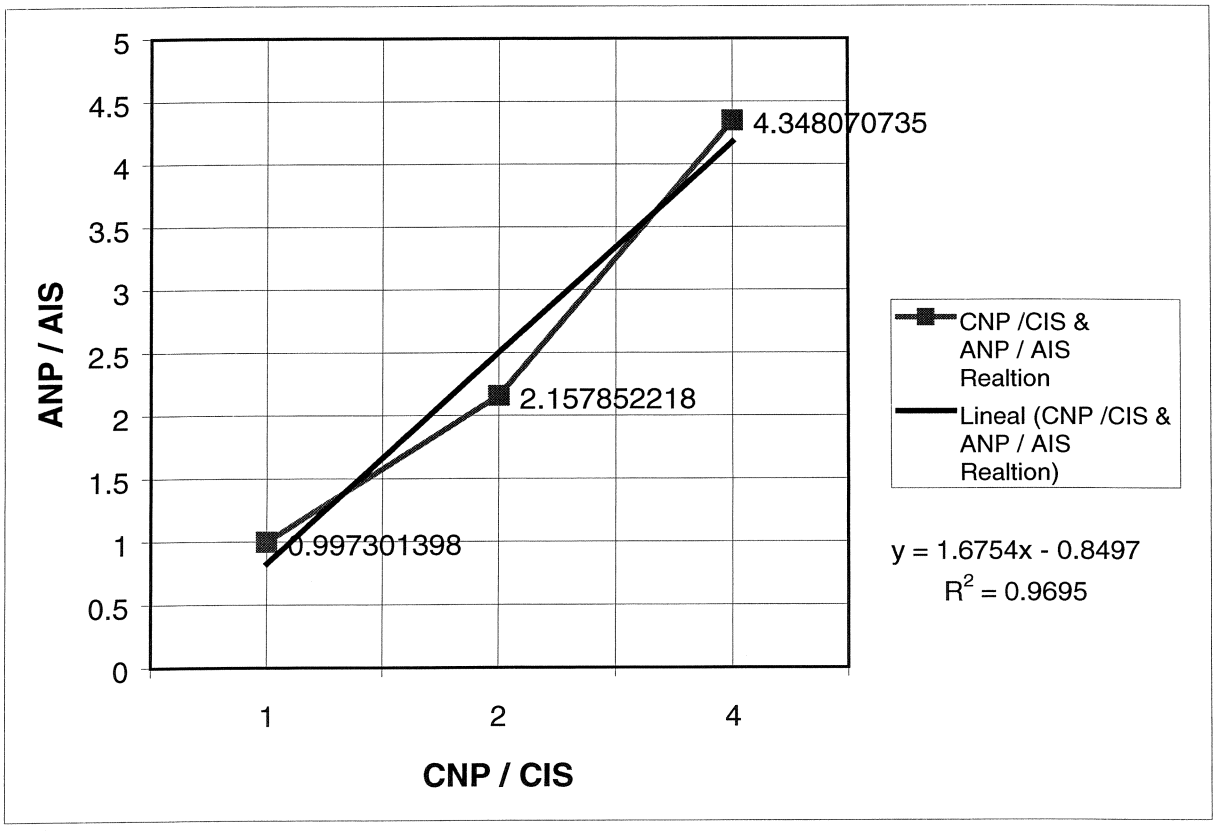


Figure 2: Calibration curve for nonylphenols in water extracts

The slope of the linearized curve was m = 1.5453 with a good regression factor: 0.9296. For the sample to be analysed is used the same graph (C_{NP} / C_{IS} vs. A_{NP} / AIS) is used with the following formula

$$\frac{C_{NP}}{C_{IS}} = \frac{\frac{W_{NP}}{V_{FINAL}}}{\frac{W_{IS}}{V_{FINAL}}}$$

Where the W_{NP} and W_{IS} are the weights of NP and IS respectively. V_{FINAL} is the final volume of the dissolution (in heptane or water) of the sample just before they were injected. So because the NP and IS were reduced until the same volume, it could be eliminated from the equation (Ullman *et al.*, 1996):

$$\frac{C_{NP}}{C_{IS}} = \frac{\frac{W_{NP}}{\cancel{V_{FINAL}}}}{\frac{W_{IS}}{\cancel{V_{FINAL}}}} = \frac{W_{NP}}{W_{IS}}$$

The graphs of W_{NP} / W_{IS} vs. A_{NP} / A_{IS} were plotted to calculate the amount of nonylphenols in the samples. The next two graphs show the relationships:

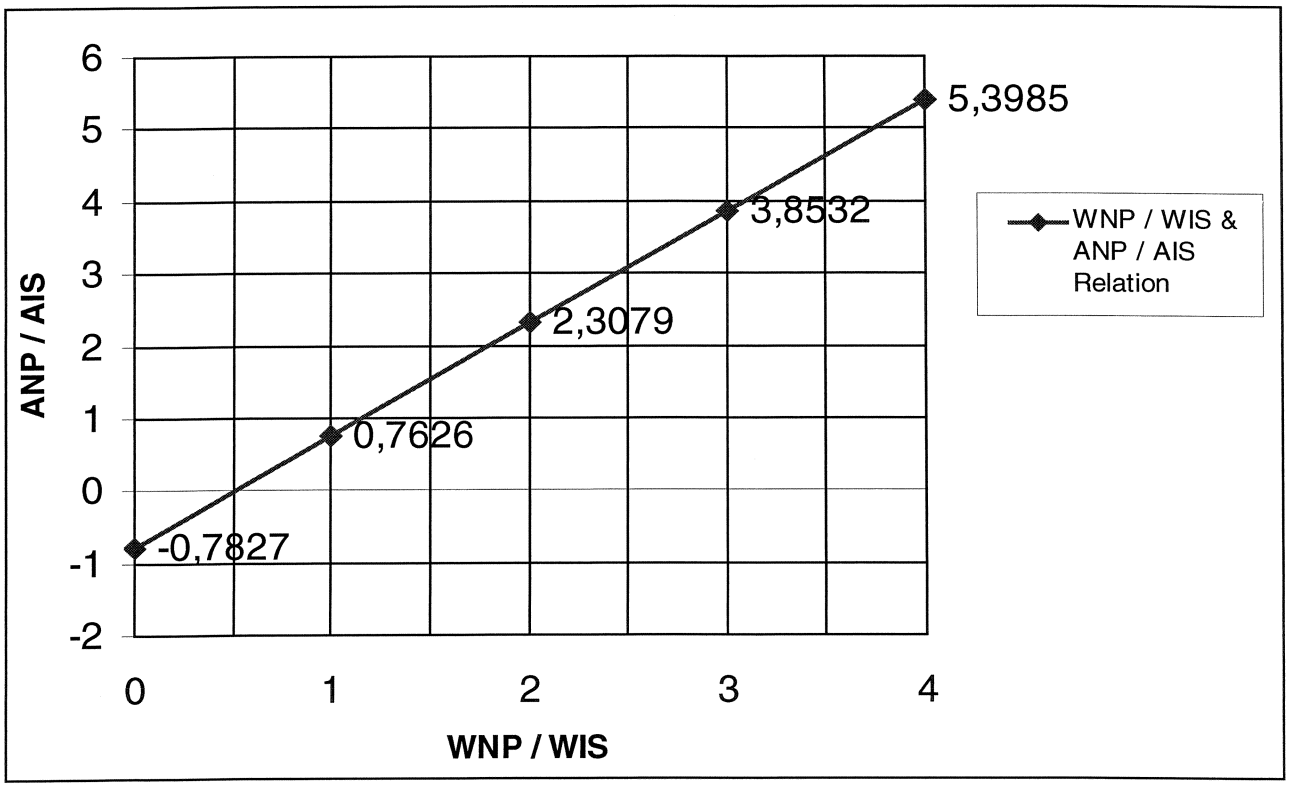


Figure 3: Relationship between WNP / WIS and ANP/ AIS in solvent extracts

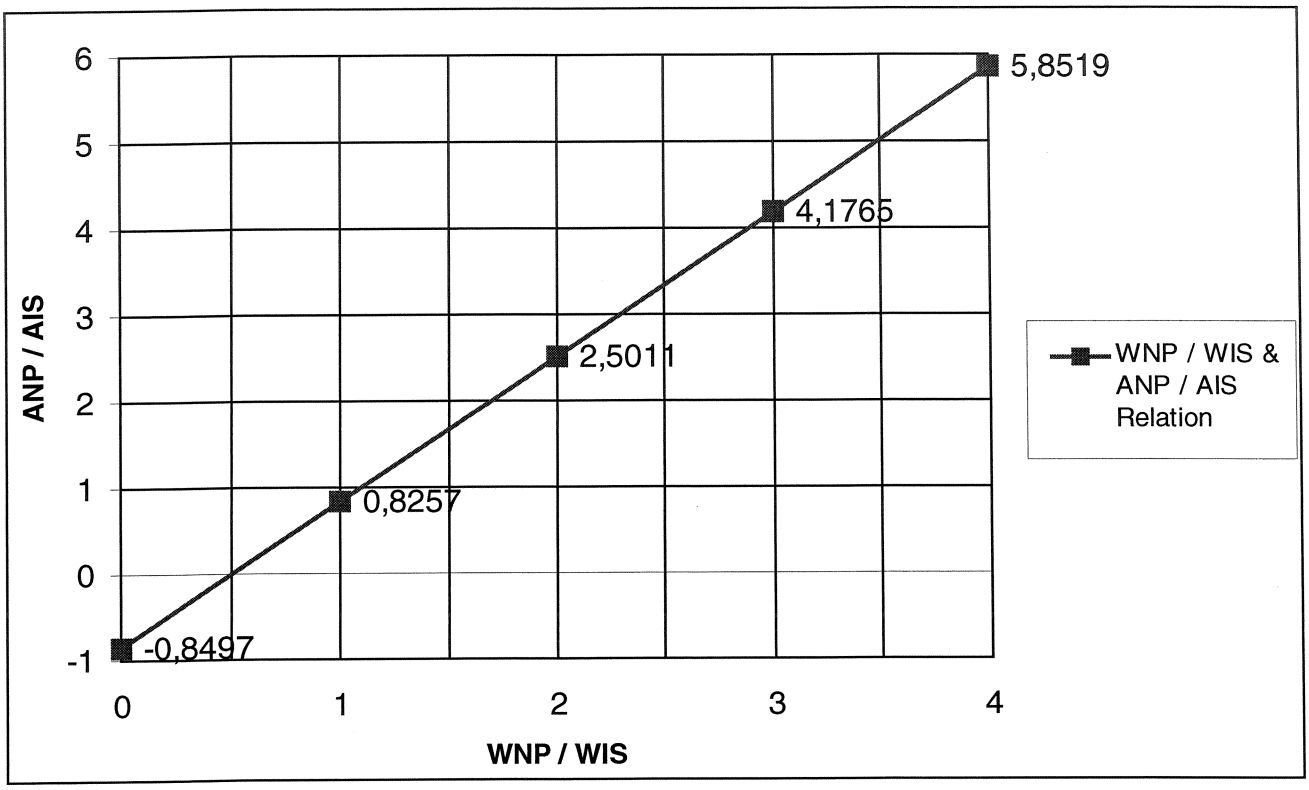


Figure 4: Relationship between WNP / WIS and ANP / AIS in water extracts

From these graphs we can say that the equation y = mx + b means now:

$A_{NP} / A_{IS} = m (W_{NP} / W_{IS}^*) + b$

Where:

A_{NP} = Total area of nonylphenols in the experimental sample
A_{IS} = Total area of internal standard in the experimental sample
m = The slope determined before for each case (Heptane or Water)
W_{NP} = Weight of nonylphenols in the experimental sample (µg)
W_{IS}^* = Weight added of internal standard in the experimental sample multiplied by the purity of the reagent (µg)
b = Ordinate to origin determined before for each case (Heptane or Water)

In order to calculate the amount of nonylphenols found in each sample (Heptane and Water) with the data and areas obtained in the chromatographic analysis, W_{NP} is in the formula:

$$W_{NP} = \frac{\frac{A_{NP}}{A_{IS}} - b}{m} \times W_{IS}^*$$

The mass of NP obtained in all the samples is shown in Table 20 – 23 in the Appendix. The amount is divided with the dry weight of the respective soil or sediment sample. The obtained value [NP] is the total amount of nonylphenols in mg per kg of dry soil or sediment. These values are shown in Tables 6 – 9 and Figure 33 – 34 in Chapter 4.