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# **Solar UV-Photooxidation as sample pretreatment step in trace metals analysis in Natural water samples by differential pulse Voltammetric methods**

## **Dissertation**

**In fulfillment of the requirement of the degree of Doctor of Philosophy in Natural Sciences (Dr. rer. nat.)**

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## **Abstract**

### **Solar UV-Photooxidation as sample pretreatment step in trace metals analysis in Natural water samples by differential pulse Voltammetric methods.**

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This dissertation describes the application of Solar ultraviolet radiation to the digestion of dissolved organic matter in natural water samples such as river water prior to the determination of trace metals by stripping voltammetric techniques. We studied that solar-UV can destroy dissolved organic matter that hinders voltammetric determination by either complexing the metal ions or by interacting with the electrode materials and optimized time required for Solar UV irradiation at a given UV intensity such as mid-latitude (Europe) or tropical regions such as Africa. We also investigated economically affordable and locally available materials and reagents required in the process of Solar UV photooxidation process.

The newly introduced protocol is applied in practical study of the determination of seven heavy metals in Awash River water (Ethiopia) by Anodic stripping voltammetric (ASV) determination of Zn, Cd, Pb, and Cu as well as in Adsorptive stripping voltammetric (AdSV) determination of trace Ni, Co and U in natural water samples. The method is clean and environmentally friendly. It can be useful in remote laboratories where electric high-power UV lamps or commercial UV digestion units are unavailable or unaffordable.

**Keywords:** Adsorptive stripping Voltammetry (AdSV), UV digestion, Pseudopolarography, River water, Nickel, Uranium.

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## List of Publications

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## List of Abbreviations

°C	Degree centigrade
μL	Micro liter
μm	micrometer
AAS	Atomic absorption spectrometry
AdSV	Adsorptive stripping voltammetry
AES	Atomic emission spectrometry
APDC	Ammonia pyrolidine dithiocarbamate
ASTU	Adama Science and Technology University
ASV	Anodic stripping voltammetry
atm	Atmosphere
ATSDR	Agency for toxic substances and disease registry
CAA	Chloranilic acid
CV	Cyclic voltammetry
D <sub>2</sub> EHPA	Diethylhexyl phosphoric acid
DAAD	Deutscher Acadamischer Austausch Dienst (German Academic Exchange Service)
DFG	German Research Foundation
DMG	Dimethylglyoxim
DNA	deoxyribonucleic acid
DO	Dissolved oxygen
DOC	organic carbon
DOM	Dissolved organic matter
Dp	Differential pulse

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DpAdSV	Differential pulse adsorptive stripping voltammetry
DpASV	Differential pulse Anodic stripping voltammetry
DPV	differential pulse voltammetry
E	Potential
$E_{acc}$	Accumulation potential
ecbp	Engineering capacity building
EPA	Environmental protection agency
$E_{sc.Inc}$	Potential scan increment
$E_{sc.rate}$	Potential scan rate
EUV	Extreme Ultraviolet
FAAS	Flame atomic absorption spectroscopy
FUV	Far Ultraviolet
GFAAS	Graphite furnace atomic absorption spectroscopy
GmbH	Gesellschaft mit beschränkter Haftung
GPES	General Purpose Electrochemical System
HCB	Hexachlorobenzene
HDV	Hydrodynamic voltammetry
HMDE	Hanging mercury drop electrode
I	Current
IC	Ion chromatography
ICP	Inductively coupled plasma
ICP-AES	Inductively coupled plasma atomic emission spectrometry
ICP-MS	Inductively coupled plasma mass spectrometry
IDL	Instrumental detection limit
IR	Infrared
ISO	International Organization for Standardization
K	Kelvin

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LSV	Linear sweep voltammetry
LUV	Low Ultraviolet
M	Moles per liter
MDL	Method detection limit
MIBK	Methyl isobutyl ketone
MUV	Middle Ultraviolet
MW	Microwave
MΩ	Mega Ohms
NHE	Normal hydrogen electrode
nm	nanometer
NPV	Normal pulse voltammetry
NUV	near Ultraviolet
Pa	Paskal
PCP	Pentachlorophenol
PET	Polyethylene terephthalate
ppb	parts per billion
ppm	parts per million
PTFE	Polytetrafluoroethane
Ref	Reference electrode
SCE	Saturated calomel electrode
SFE	Supercritical Fluid Extraction
SMDE	Static mercury drop electrode
SODIS	Solar water disinfection
SoUV	Solar ultraviolet
SUV	Super Ultraviolet
SWV	Square wave voltammetry
TEA	Triethanolamine

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$t_{\text{int}}$	Time interval
TMFE	Thin mercury film electrode
TNT	1, 3, 5,-trinitrotoluene
TOC	Total organic carbon
USEPA	United States Environmental protection agency
UV	Ultraviolet
UVA	Ultraviolet A
UVB	Ultraviolet B
UVC	Ultraviolet C
UV-VIS	Ultraviolet-Visible
V	Voltage
VA	Voltammetry
VUV	Vacuum Ultraviolet
W	Watt
WHO	World health organization
XRF	x-ray fluorescence
$\lambda$	Lambda(wavelength
$\nu$	nu (Frequency)



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## 1. Introduction

Sample Preparation or pretreatment step is the most crucial step in any analysis to obtain a representative and homogeneous sample. In this level we may need to pass through several steps depending on the nature of the sample, the type of the analytes, the type of the instrument and the method we employ for the determination. Sample preparation minimizes problems related to homogeneity, representativity, time consumption and handling steps [Capelo-Martinez 2000]. Solid samples have to be converted to liquids by decomposition. This will destroy the matrix and separates interfering substances. Liquid samples have to be treated in a way that the analyte species is separated from the matrix to make it available for determination. This will facilitate the isoformation of samples and standards, homogenization and pre-concentration of analytes. Sample pretreatment must be analytically accurate with no or minimum contamination, minimum loss of elements, complete decomposition, small number of working steps and reliable equipment [Mitra 2003]. It has to be economically efficient (low consumption of chemicals, ease of handling, low investment costs, low operating costs and automation). Sample preparation step should be safe and easy to perform. Sample digestion influences analysis time and it is an important economic factor in the analytical laboratories. The determination of trace metals in natural waters like river water, sea water, Lake, etc is crucial especially for those metals causing health hazards and environmental effects. Metal pollutants exist bounded by organic and/ or inorganic matrices in the environment and thus the determination of these metals in natural waters becomes complicated unless they are separated from the matrices by application of proper sample pretreatment methods [Achterberg 2001]. Electrochemical methods especially Voltammetric determination of metal ions requires homogenous samples, free of organic matter which interacts with the metal ions and with the electrode material. Dissolved organic matter (DOM) may form complexes with metal ions preventing them from reduction at the electrodes or shifting the electrochemical redox potential and also hinders the determination by interacting with the electrode material or undergo electrochemical redox reaction at the electrodes resulting increase of the background current and making the determination impossible [Golimowski 1996]. Methods like wet digestion and dry ashing have been introduced long ago in the mineralization of organic and inorganic matter for both liquid and solid samples. Application of these methods is associated with high risk of contamination of the sample due to the addition of different reagents like mineral acids, bisulfate ions, and other oxidants. They

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require ultra pure reagents which is relatively unaffordable by many laboratories especially in developing countries. To avoid problems related to contamination, cost and safety due to the introduction of corrosive reagents like mineral acids and oxidants, an alternative clean and efficient method of sample treatment method has been developed long ago [Armstrong 1966, Achterberg 1994]. This new method of sample pretreatment method is Ultraviolet (UV) digestion method. The method is considered environmentally friendly, effective and minimizes the risks of contamination or loss of sample through evaporation [Achterberg 1994]. UV radiation effects the formation of different reactive radicals and oxidants like singlet oxygen [Merkel 1972], superoxide radicals and alkylperoxy radicals [Baxter 1983], hydroxyl radical and hydrogen peroxide [Zhou X 1990] are formed. The subsequent reaction of these radicals with organic matter was known to be one of the natural ways of biodegradation in aquatic system [Mill 1990]. UV radiation generated from artificial UV sources have been employed in the decomposition of (1,1(4,4'-dichlorodiphenyl)2,2,2-trichloroethane), HCB (hexachlorobenzene), PCP (pentachlorophenol), TNT (1,3,5-trinitrotoluene) [Symp. Pap.-Heraeus 1990], atrazine (a herbicide) [Vieweg T. 1992] and others decompose following absorption of 180-250 nm radiation. Microwave-assisted, high-temperature UV digestion procedure was developed for the accelerated decomposition of interfering dissolved organic carbon (DOC) prior to trace element analysis of liquid samples such as, industrial/municipal wastewater, groundwater, and surface water, body fluids, infusions, beverages, and sewage [Wegelin 2001]. But these instruments are relatively expensive and make them difficult to be used in some laboratories. Solar UV is increasingly investigated in the areas of photochemistry and biophotochemistry, because of its prominent ecological and environmental roles, cost effectiveness and safety matters. In addition to biological activities, solar UV is the most important element in natural disintegration of most organic compounds in soil, water, and air [Al-Rasheed R. 2003]. Direct Solar UV radiation in combination with Titania ( $\text{TiO}_2$ ) photocatalysts was tested for its application to the disinfection of water and destruction of pathogens and mineralization of organic compounds [Minero 1996. Robert 2002]. Humic acids, fulvic acids, glycolic acid, peptides, proteins, aminoacids, lipids, and polysaccharides are the most dominant organic constituents of natural waters like River, lake, sea water [Uyguner 2005]. Humic acids for instance, contain negatively charged ions due to the dissociation of carboxylic acid and hydroxyl functional groups and also possess amphipathic character owing to the presence of both hydrophobic and hydrophilic moieties [Eggins 1997]. These characteristic of humic acid enables it to bind

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metal ions hindering from reduction at the electrode or by interacting themselves with the electrode surface. The destruction of these organic matters from the sample is unquestioned.

### ***1.1. Objectives***

This doctoral research is aimed to achieve the following objective.

- (a) To develop a new, affordable, environmentally friendly, effective and efficient method of sample treatment method prior to voltammetric determination of trace metals in natural waters.
- (b) To investigate the applicability of Solar-UV photooxidation as sample pretreatment step in the determination of trace metals by differential pulse anodic and adsorptive stripping voltammetric methods.
- (c) To analyze the degree of contamination of Awash River in respect to some heavy metals by applying the newly developed sample pretreatment method

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## **2. Theoretical Background**

### **2.1. Sample Preparation Methods for metals analysis**

Prior to performance of any analysis, it is important to obtain a representative and homogeneous sample via appropriate treatment as required for specified analytical method. Sample preparation is frequently a major time-consuming step in most analyses because it is rarely possible to analyze a neat sample. In sample preparation step we may pass through mechanical sample preparation steps such as grinding, drying, weighing and so forth. It is also important to transfer a solid or liquid sample into a soluble or dissolved state for subsequent analytical characterization of its major, minor and trace constituents in the decomposition steps [Sures 1995]. Digestion is a method of converting the complex components of a matrix into simple chemical forms by supplying energy such as heat and microwave energies or by adding chemical reagents such as acids or a combination of the two methods. The digested sample will be available for trace metals determination by various spectroscopic, chromatographic or electrochemical methods such as FAAS or GFAAS, AES, ICP-AES, ICPMS, XRF, IC and (VA)[Golimowski 1975, Sipos, 1979, Yusof 1994]. A common result of the sample preparation is the dissolution of the entire sample, producing a clear solution. Digestion methods will be developed based on the nature of the sample, nature of the matrix, the analyte, analysis instrument (technique) and determination extent of which the digestion is needed. The nature of matrix, analyte, reaction properties and instrumentation can influence the extent and type of decomposition to a larger extent. The nature of the matrix influences its reactivity, homogeneity and sample size. The nature of analyte influences the elements of interest, concentration, molecular interaction, analyte final form and instrumental compatibility. Reaction properties influence reagent interaction at specific temperature, solubility and volatility of analyte and matrix components. Instrumentation determines the type of instrument used, sample throughput, detection limit, sample introduction system and clean chemistry facilities [Jage 1992]. A number of decomposition or digestion methods are developed both for total mineralization or leaching purposes. Among these, most important ones are discussed in the next sections.

#### **2.1.1. Wet Digestion Methods**

Wet digestion is sometimes referred interchangeably as decomposition, dissolution, destruction, digestion, wet ashing, mineralization or oxidative acid digestion. Wet digestion is

the most common and conventional Sample preparation method used for dissolving samples for metals analysis. The digestion can take place in an open flask or in a pressurized (sealed) container by heating with conventional method or with microwave oven. In this method, samples to be analyzed for elemental metal content are usually prepared by digesting the matrix in a strong acid. In the case of organic matrices, an oxidizing mixture is used to destroy the entire organic matrix and solubilize the sample to a clear solution containing the metals available for analysis by such techniques as VA, AAS, ICP, or ICP-MS. The first published digestion reagent was chloric acid HCl + KCl [Duflos 1938]. The most effective classical wet digestion mixtures (reagents) are HNO<sub>3</sub> + H<sub>2</sub>SO<sub>4</sub> are used for the destruction of organic matter. The majority of wet digestion methods ( total destruction) involves the use of some combination of oxidizing acids(HNO<sub>3</sub>, hot conc. HClO<sub>4</sub>, hot conc. H<sub>2</sub>SO<sub>4</sub>) and non oxidizing acids(HCl, HF, H<sub>3</sub>PO<sub>4</sub>, dil. H<sub>2</sub>SO<sub>4</sub>, dil. HClO<sub>4</sub>) and hydrogen peroxide. Nitric acid is commonly used with or without Oxidant (e.g. hydrogen peroxide), because there is no chance of forming insoluble salts as might happen with HCl or H<sub>2</sub>SO<sub>4</sub>. Inorganic samples, soils, sediments, ores, rocks, and minerals may be digested in dilute or concentrated acids or mixtures of acids, which may be sufficient to leach out the analytes (table 2.1). Hydrofluoric acid can be used if total dissolution is required. Refractory materials such as cements, ceramics, and glass may require fusion or flux digestion, which involves melting the ground sample with a salt such as sodium carbonate or sodium peroxide. However, the method is not generally suitable for metals, which tend to be lost by volatilization because of the high temperatures required. In addition, the material of the container becomes more critical as the aggressiveness of the digestion process increases [Mitra 2003].

**Table 2.1:** Digestion reagents and their application in wet digestion

<b>Reagent</b>	<b>Sample type</b>
Water	Soluble salts
Dilute acids	Dry-ashed sample residues, easily oxidized metals and alloys, salts
Concentrated acid (e.g., HNO <sub>3</sub> )	Less readily oxidized metals and alloys, steels, metal oxides
Concentrated acid with added oxidizing agent (eg.H <sub>2</sub> O <sub>2</sub> )	Metals, alloys, soils, particulates from air, refractory minerals, vegetable matter
Hydrofluoric acid	Silicates and other rock samples

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The simplest method for acid digestion or wet-ashing is carried out in an open container in which samples are dried, weighed, and placed in a beaker in the presence of digestion reagent. Acid digestion in both open and closed (pressurized) systems is carried out with the aid of a variety of reagents such as mineral acids. Other reagents like hydrogen peroxide, potassium peroxide sulfate, boric acid and many more are employed depending on the type of the sample to be digested [Mitra 2003]. The sample is allowed to boil very gently until it has been digested completely and it will be evaporated to near dryness and then taken up in a dilute acid solution and diluted to volume for analysis. Open systems can utilize hot plates or block techniques, microwave heating or UV- digestion. Simple equipment high number of samples and large sample weight is the advantage of open system. However a number of short comings are associated with this method. Some of these short comings are high reagent consumption, reagent bulk, risk of contamination, evaporation (loss) of analyte, limited temperature and long decomposition time. Wet Ashing in a closed system is sometimes termed as pressure ashing and it applies similar methods such as conventional heating, microwave and UV digestion [Kotz 1974]. In this method the weighed samples are placed into small quartz vessels with the appropriate acid digestion solution sealed with PTFE and quartz caps, placed in a heating block, and the apparatus closed and pressurized with nitrogen [Welz 1999]. The nitrogen serves to support the digestion vessels by equalizing pressure inside and outside the vessels and to introduce inert environment in the digestion vessel. A number of advantages of this system can be mentioned. It allows typically high temperature(200-260°C and this results in dramatic increase in reaction kinetics allowing digestion to be carried out in a matter of hours in addition to short reaction time, no volatilization (loss of analyte) occurs. Similarly less reagent consumption and less contamination could be achieved. A limited sample weight and safety hazard due to high pressure can be taken as limitations of this system.

### **2.1.2. Microwave-assisted wet Digestion**

Microwave energy belongs to non-classical energy sources together with ultrasound, high pressure, mechanical activation, or plasma discharge [Nüchter 2000]. Microwave activation increases efficiencies of many chemical processes at the same time as it can reduce formation of by-products, otherwise obtained from conventionally heated reactions. Since the first reports on the use of microwave heating to accelerate organic chemical transformations [Abu-Samra 1975, Gedye 1986, Giguere 1986], thousands of articles have been published in the area of microwave-assisted synthesis and sample treatment topics. It has been applied in the

digestion of geological, biological, clinical, botanical, food, environmental, sludge, coal and ash, metallic, synthetic and mixed samples. In most of the MW processes, the digestions are carried out in closed containers made up of chemically inert and MW transparent materials such as polycarbonates and PTFE which are less likely to contain metal contaminants than are glass or ceramic beakers or crucibles. There are two types of microwave digestion systems, focused (open) and Pressurized (closed). Subsequent research showed that using focused(open) microwave digestion produces equivalent results to those of traditional heating techniques but in much shorter time [Jones 1918]. Digesting a sample in a closed container in a MW oven has several advantages over open container dissolution methods. The sealed container eliminates the chance of airborne dust contamination. The closed, pressurized containers reduce evaporation, so that less acid digestion solution is required, reducing blanks. The sealed container also eliminates losses of more volatile metal species, which can be a problem in open container sample decomposition, especially in dry ashing. In general, the use of MW digestion is preferable for practical reasons. Microwave energy is delivered into the sample efficiently without heating containers, hotplates, and so on. The energy can readily be controlled and programmed automatically, ensuring better reproducibility. Sample digestion times are reduced significantly, and the amount of reagent required is usually less. Additionally, there is less chance of volatilization of some analytes, and sample contamination is less likely than when an open container is used. Finally, microwaves provide an excellent opportunity for automation [Parr 2001].

**Table 2.2: Wet digestion scheme**

System	Digestion technique	Reagents	Application
Open system	Conventional heating	HNO <sub>3</sub> , HCl, HF, H <sub>2</sub> SO <sub>4</sub> , HClO <sub>4</sub> , H <sub>2</sub> O <sub>2</sub>	Organic/Inorganic
	Microwave heating	HNO <sub>3</sub> , HCl, HF, H <sub>2</sub> SO <sub>4</sub> , HClO <sub>4</sub> , H <sub>2</sub> O <sub>2</sub>	Organic/Inorganic
	Ultraviolet heating	H <sub>2</sub> O <sub>2</sub> , K <sub>2</sub> S <sub>2</sub> O <sub>8</sub>	Water, Slurries
Closed system	Conventional heating	HNO <sub>3</sub> , HCl, HF, H <sub>2</sub> O <sub>2</sub>	Organic/Inorganic
	Microwave heating	HNO <sub>3</sub> , HCl, HF, H <sub>2</sub> O <sub>2</sub>	Organic/Inorganic
Flow system	Conventional heating	HNO <sub>3</sub> , H <sub>2</sub> SO <sub>4</sub> , H <sub>2</sub> O <sub>2</sub>	Organic/Inorganic
	UV On-line decomposition	H <sub>2</sub> O <sub>2</sub> , K <sub>2</sub> S <sub>2</sub> O <sub>8</sub>	Water, Slurries
	Microwave heating	HNO <sub>3</sub> , H <sub>2</sub> SO <sub>4</sub> , H <sub>2</sub> O <sub>2</sub>	Organic/Inorganic
Vapor-phase acid digestion		HNO <sub>3</sub> , HCl, HF, H <sub>2</sub> O <sub>2</sub>	Organic/Inorganic

### 2.1.3. Dry Ashing method

Dry Ashing is performed by placing the sample in an open inert vessel and destroying the combustible (organic) portion of the sample by thermal decomposition using a muffle furnace

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[Friel 1986]. Ashing temperatures usually range from 450 to 550°C. Additives such as sulfuric acid or salts such as Magnesium nitrate may aid in dry ashing. Sulfuric acid has a chemical charring effect, and salts such as magnesium nitrate, sodium carbonate, and magnesium oxide aid in the retention of some elements. These salts leave a soluble alkaline inorganic residue [Tsalev 1983]. Silica remaining after destruction of much of the sample matrix can occlude metals and render them insoluble in the acid used to dissolve the residue. If this is a major difficulty with certain samples, further treatment with hydrofluoric acid may be needed to dissolve the silica entirely [Isaac 1975]. Usually, the sample is charred using an open flame (crucibles/clay triangles/Bunsen and/or Meeker burners under a hood or by addition of sulfuric acid prior to muffle). Dry ashing is a relatively simple method of removing the organic matter that can be used for relatively large samples and requires little of the analyst's time. Dry ashing is suitable for nutritional elements in foods, such as Fe, K, Ca, Mg, and Mn, which are present in substantial quantity and are stable at the high temperatures required [Mitra 2003]. The major drawbacks of the method are the possible loss of some elements by volatilization, contamination of the sample by airborne dust, as it must be left open to the atmosphere and irreversible sorption of analyte into the walls of the vessel. However, losses from volatilization of the analyte can be minimized by restricting the temperature at which ashing takes place. For determination of lead, copper, zinc, cadmium, and iron in foodstuffs, for example, good recoveries of the analytes were obtained by heating the samples slowly to 450°C and holding this temperature for 1 hour [Jorhem 2000].

#### **2.1.4. Organic Extraction of Metals**

Solvent extraction and partitioning, is a method to separate compounds based on their relative solubilities in two different immiscible liquids, usually water and an organic solvent. It is an extraction of a substance from one liquid phase into another liquid phase. Liquid–liquid extraction is a basic technique in chemical laboratories, where it is performed using a separatory funnel. Ionic species, including metallic ions, are quite insoluble in organic Solvents. If the charge on the metal ion is neutralized or the ion is bound to a larger organic moiety, the metal becomes soluble in an organic solvent and, consequently, can be extracted from the aqueous phase. This can be achieved either by formation of metal chelates, metal–organic complexes, or by ion pairing [Tessier 1979]. A complex formed between a metal and a chelating agent is hydrophobic in nature and soluble in organic solvents. Therefore Organic extraction is carried out for recovery of dissolved metals from water samples. The most



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common chelating agents used to extract metals from water samples are ammonia pyroline dithiocarbamate (APDC) and 8-hydroxyquinone are generally used as a solvent. The other important commercial liquid-liquid extraction systems for some metals like copper, cobalt and nickel involve contacting an aqueous acid phase with some organic acid (HR)[Hughes 1984]. cobalt can be extracted from hydrochloric acid using alamine 336 in *m*-xylene or by using cyanex 272 (*bis*-(2,4,4-trimethylpentyl)phosphinic acid[Mackenzie 2008]. Nickel can be extracted using di(2-ethyl-hexyl)phosphoric acid and tributyl phosphate in a hydrocarbon diluent (Shellsol). The zinc and cadmium are both extracted by an ion exchange process, the *N,N,N',N'*-tetrakis(2-pyridylmethyl)ethylenediamine (TPEN) acts as a masking agent for the zinc and an extractant for the cadmium[Takeshita K 2003]. In the modified Zincex process, zinc is separated from most divalent ions by solvent extraction. D<sub>2</sub>EHPA (Diethyl hexyl phosphoric acid) is used for this. A zinc ion replaces the proton from two D<sub>2</sub>EHPA molecules. To strip the zinc from the D<sub>2</sub>EHPA, sulfuric acid is used, at a concentration of above 170 g.L<sup>-1</sup> (typically 240-265g.L<sup>-1</sup>). According to Halwachs precious metal chemistry and chemical engineering (2008), Dialkyl sulfides, tributyl phosphate and alkyl amines have been used for extracting palladium and platinum.

### **2.1.5. Extraction with Supercritical Fluids**

Supercritical Fluid Extraction (SFE) is the process of separating one component (the extract ant) from another (the matrix) using supercritical fluids as the extracting solvent. It is a method of extracting a metal or metalloid species from a solid or liquid material by exposing the material to a supercritical fluid containing chelating agent [McHugh 1986]. Supercritical Fluid Extraction can be used as a sample preparation step for analytical purposes, or on a larger scale to either strip unwanted material from a product (e.g. decaffeination) or collect a desired product (e.g. essential oils). Carbon dioxide (CO<sub>2</sub>) is the most used supercritical fluid, sometimes modified by co-solvents such as ethanol or methanol. It is an alternative to liquid extraction using solvents such as hexane or dichloromethane. The properties of a supercritical fluid can be altered by varying the pressure and temperature, allowing selective extraction. For example, volatile oils can be extracted from a plant with low pressures (100 bar), whereas liquid extraction would also remove lipids. Lipids can be removed using pure CO<sub>2</sub> at higher pressures, and then phospholipids can be removed by adding ethanol to the solvent [Tanaka 2007]. Solvent evaporation and disposal are eliminated, and the extractions may be very efficient because of the low viscosity of supercritical fluids, which allows them to penetrate

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readily into the solid sample particles. Carbon dioxide, with or without modifiers such as methanol, is the most commonly used solvent. For extraction with supercritical carbon dioxide, metals are first chelated with a ligand such as a derivative of dithiocarbamate. It has been found that while the solubility of chelates of metals with sodium diethyl dithiocarbamate in carbon dioxide is quite low. The solubility can be increased significantly by substitution of a longer chain alkyl group for the ethyl groups on the dithiocarbamate. Even better extractions were obtained when the ethyl groups of the diethyl dithiocarbamate were fluorinated.

### **2.1.6. Ultrasonic Sample Preparation**

Some sample matrices are inherently difficult to ash. Foodstuffs with high sugar content are good examples. Dry ashing must be done slowly and requires over 30 hours [Luque de Castro 2007]. Some Soluble samples such as sugar can be analyzed directly or after dilution to a desired extent to obtain high detection limits, and the recovery of analytes tends to be low. To avoid dilution problem, an extraction method that uses ultrasonic probe has been developed [Chimilenko 1998]. The sample (example sugar) is mixed with water and is ultrasonicated for a period of time to ensure thorough solution. Then the pH is adjusted to appropriate level and with addition of aqueous reagent like sodium diethyl dithiocarbamate. Extraction of the solution is carried out with chloroform at least twice. The extract is evaporated and the residue is taken up in dilute acid for analysis [Mitra 2003]. Ultrasonication for soil samples in 1:1 diluted aqua regia was found to give excellent recovery of As, Cd, Pb, and Ag from reference samples. The results were comparable to those obtained by microwave digestion, and the speed of extraction and sample throughput was better with the ultrasonication [Vaisanen 2002].

### **2.1.7. Ultraviolet (UV) Sample Preparation methods**

#### **a. UV-Radiation and UV-Radiation Sources**

Ultraviolet (UV) light is electromagnetic radiation with a wave length ranging from 10 nm to 400 nm with energies between 3eV and 124eV. UV radiation has a wave length shorter than visible light and longer than X-Ray. It was discovered when the German physicist Joann Wilhelm Ritter observed that, invisible rays just beyond the violet end of the visible spectrum darkened silver chloride-soaked paper quicker than the violet light itself. He called them "oxidizing rays" until the term is replaces by Ultraviolet in favor of the violet light [Steven 1989, Hockberger 2002]. According to ISO standard on determining solar irradiance

(ISODIS- 21348), the electromagnetic spectrum of ultraviolet light is described as shown in table 2.3. The main natural source of UV is the sun. Extreme UV, UVC, and UVB constitute 99% of the total UV radiation reaching the planet Earth and are absorbed in the upper atmosphere by ozone, oxygen, and nitrogen [Philip 2001]. The atmosphere is transparent to UVA (315– 400 nm), and 99% of the total UV-radiation reaching the earth’s crust is UVA. This radiation may reach the ground scattered, diffused, or reflected. Its intensity is also dependent on the cloudiness, latitude, season, altitude, elevation of the sun, and so forth.

**Table 2.3:** Classification of ultraviolet light based on International Organization for standardization (ISO)

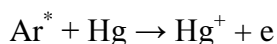
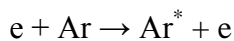
Name	Abbreviation	Wavelength Range(nm)	Energy (eV)
Ultraviolet A	UVA	400 - 315	3.10 – 3.94
Near UV	NUV	400 - 300	3.10 – 4.13
Ultraviolet B or medium wave	UVB	315 - 280	3.94 – 4.43
Middle UV	MUV	300 - 200	4.13 – 6.20
Ultraviolet C	UVC	280 - 100	4.43 – 12.4
Far UV	FUV	200 - 122	6.20 – 10.2
Vacuum UV	VUV	200 - 100	6.20 – 12.4
Low UV	LUV	100 - 88	12.4 – 14.1
Super UV	SUV	150 - 10	8.28 – 124
Extreme UV	EUV	121 - 10	10.2 – 124

Transmittance is also dependent on the optical property of the transparent object, or it is a function of the type, thickness, angle of incidence, and specific wavelength of the materials [Postupolski 1991]. Identification and optimization of the transmittance of large number of materials has been investigated. Pyrex glass (borosilicate type) has maximum transmission level at 340 nm and beyond [Acra 1980]. Transparent plastic materials include Lucite and Plexiglass [Dietz 1963], polyethylene [Fujioka 1982], and polyethylene terephthalate (PET) [Adler 1991]. In case of water, transmittance depends on the water depth and turbidity, which internally depends on the presence or absence of light-absorbing and coloring materials, and mineral salts, humates, as well as wavelength of the radiation [Calkins 1974, Gameson 1967, Ogura 1969, Qualls 1983]. There are different artificial UV sources of which mercury lamp is the most frequently used. Although the mercury spectrum is rich in lines in the UV range (100-600 nm), the resonance line or the transition energy of Hg atom from lowest excited state to the ground state corresponds to 253.7 nm. Also Mercury is a relatively inert element which neither reacts with the electrode material nor with the bulb material especially in the

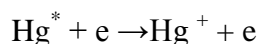
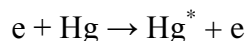
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inert atmosphere like Argon. R. Phillips [Phillips 1983] described that Argon participates in photon generation indirectly by increasing the number of current carrying electrons and ions as described below Lamp ignition step Lamp operation step.

**Lamp ignition step**



**Lamp operation step**



Most often, Mercury lamps are divided into 2 groups: low pressure lamps (Hg pressure 0.1-1.0 Pa ( $10^{-3}$ - $10^{-2}$  Torr) and high-pressure lamps (Hg pressure > 0.1 MPa (760 torr). Low pressure lamp is a glass tube of about 1 m long and 15- 40 mm diameter containing a mixture of Hg vapor (0.1-1Pa) and a noble gas, predominantly Argon at about 100 Pa pressure. The spectrum of a low-pressure lamp consists of almost only 2 lines: the resonance line at 253.7 nm of the  $6^3P_1 \rightarrow 6^1S_0$  transition and the line at 184.9 nm corresponding to the  $6^1P_1 \rightarrow 6^1S_0$  transition with the ratio of the 253.7 nm line intensity to the 184.9 nm line intensity equals 0.34 - 0.12 depending on the bulb Temperature and the arc current. These lamps have a long operating time of 5000-10000 hours and their maximal power is 60 W [Phillips 1983]. Black light fluorescent lamps are special, a type of low-pressure lamps which absorbs the radiation of 254 nm and emits the radiation of around 365 nm (also known as black-light). These types of lamps are often applied for photocatalytic oxidation of organic substances in solution with titanium dioxide as a catalyst. Similarly it can be applied in the flow-injection determination of TOC in a borosilicate apparatus [Matthews 1987, Matthews 1990]. Medium and high pressure lamps are shorter than low-pressure lamps with about 10-150 cm long and 10-40 mm in diameter. Medium-pressure lamps operate in the state of the so-called local thermal equilibrium, or the temperature of atoms and ions equals the temperature of electrons which is 5000-8000 K. The spectrum of a medium-pressure lamp is richer than the spectrum of a low pressure. In addition to the single emission lines, it also contains an emission continuum, especially in the UVC range. The contributions to each UV range in relation to the energy consumed by the lamp torch are 16% UVC, 7% UVB and 7% UVA [Symp. Pap.-Heraeus 1990]. 15% of the power is converted into visible radiation and 35% into thermal radiation (IR and lamp walls radiation). The remaining 20% is lost in the process of heat conduction on the electrodes [Golimowski 1996]. The material of the lamp bulb must fulfill many conditions, Such as UV radiation transmittance, chemical resistance to the substances contained in a lamp, strength at elevated temperature and pressure.

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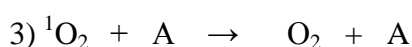
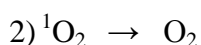
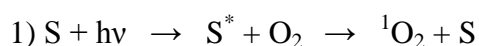
**b. Types and Mechanisms of formation of Oxidants under the influence of UV Radiation and organic matter destruction**

UV radiation is capable of damaging biological organisms (example “germicidal lamps). It is also well known to destroy organic matter. Fortunately high energy UV photons such as UVC and VUB are almost completely absorbed by ozone, oxygen and other gases and only very few of them reach the earth’s surface. UVB radiation (280- 315 nm) is only partially absorbed by the ozone layer and can damage biological organisms while UVA (>315 nm) is not absorbed by ozone and generally it does not damage biological organisms. UV radiations generated from mercury lamp is used for water disinfection, sterilization of various instruments and objects, treatment of water and waste water [Koller 1965, Acra 1980]. Non biodegradable compounds such as DDT (1,1(4,4’-dichlorodiphenyl) 2,2,2-trichloroethane), HCB (hexachlorobenzene), PCP (pentachlorophenol), TNT (1,3,5,-trinitrotoluene) are known to be destroyed by UV and the mechanisms and the products of UV decompositions have been studied elsewhere [Symp. Pap.-Heraeus 1990]. Natural UV-Photooxidation is observed and investigated long ago and similar processes are applied in industrial decomposition of toxic substances, in treatment of urban and industrial waste waters and in production of ultrapure water for trace analysis. Several intermediate compounds are formed in the interaction of UV radiation with dissolved organic matter (DOM). This include excited states of DOM, hydrogen peroxide, singlet oxygen, hydrated electrons, superoxide ions, organoperoxy radicals, hydroxyl radicals and halogen radicals [Zhou 1990].

**Singlet Oxygen ( $^1\text{O}_2$ ):** Singlet oxygen ( $^1\text{O}_2$ ) is a higher energy state molecular oxygen species. It is one of the most active intermediates involved in chemical and biochemical reactions [Halliwell 1982, Ameta 1990]. It was first observed in 1924 [Wasserman 1979] and then defined as a more reactive form of oxygen [Halliwell 1982] produced by photo sensitization reaction in the presence of photo sensitizers [Krinsky 1979]. As a result of the action of UV radiation on molecular oxygen, it changes its energy state from the ground (triplet) state to the excited (singlet)  $^1\text{O}_2$  state [Merkel 1972], which is probably generated at  $\lambda = 366 \text{ nm}$  [Qualls 1983]. Being important intermediate species in the detrimental oxidation of biomolecules, it has been demonstrated that  $^1\text{O}_2$  can react with many kinds of biological molecules such as DNA, proteins and lipids [Briviba 1997]. The reactions of  $^1\text{O}_2$  often involve carbon-carbon double bond, which are present in many biological and other organic molecules, such as carotene, chlorophyll, unsaturated fatty acids, humic substances and so forth [Ameta 1990]. The first oxidation mechanism of singlet oxygen in the natural

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environment was first proposed by Kautsky [Kautsky 1939]. According to this proposal, the energy of sunlight becomes absorbed by the sensitizers, then it is transferred to the O<sub>2</sub> molecules and the molecules change their energy state to <sup>1</sup>O<sub>2</sub>. The formed singlet oxygen atoms react with water and the organic substances present forming peroxides and radicals. Haag and Hoigne' [Haag 1986] proposed the following scheme describing possible processes that occur during UV-energy.

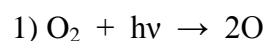


Where, S and S\* are the sensitizer e.g. humic acid in the ground and excited state, respectively, and A is the energy acceptor compound (trapping agent). Reaction (1) shows how the sensitizer participates in the process, reaction (2) shows physical quenching of excited oxygen molecules by water, reaction (3) shows physical quenching of excited oxygen molecules by an organic acceptor e.g. furfuryl alcohol and reaction 4 shows the chemical reaction with A. The addition of chemical compounds like dyes like rose bengal or methylene blue which can act like natural sensitizers and the subsequent exposure to sunlight is recommended as one of the methods of waste water treatment[Haag 1985]. Singlet oxygen has relatively short life time (2μs) in aqueous solution and its steady state concentration [<sup>1</sup>O<sub>2</sub>]<sub>ss</sub> is ca 10<sup>-12</sup> mole L<sup>-1</sup> [Ameta S.C 1990]. It was shown that there is a linear relationship between this concentration and the concentration of dissolved organic carbon (DOC), confirming the already shown mechanism of formation of the singlet oxygen [Haag 1985].

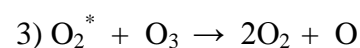
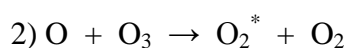
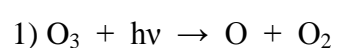
**Superoxide Radicals (O<sub>2</sub><sup>-</sup>)** Oxygen centered free radicals contain two unpaired electrons in the outer shell. The most common reactive oxygen species (ROS) include: the superoxide anion (O<sub>2</sub><sup>-</sup>), the hydroxyl radical (OH<sup>•</sup>), singlet oxygen (<sup>1</sup>O<sub>2</sub>), and hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>). According to Goldfarb A.H.[Goldfarb 1999], Superoxide anions are formed when oxygen (O<sub>2</sub>) acquires an additional electron, leaving the molecule with only one unpaired electron. Reaction takes place when free radicals take an electron from a surrounding compound or molecule generating new free radical in its place. In turn the newly formed radical then looks to return to its ground state by taking electrons with antiparallel spins from molecular or cellular structures. Thus the chain reaction continues and can be "thousand of events long. A

superoxide ion is the first product in the reduction of oxygen molecule and it is unstable in aqueous solutions and quickly disproportionate to H<sub>2</sub>O<sub>2</sub> and O<sub>2</sub> [Baxter 1983]. J. Golimowski and K. Golimowska [Golimowski 1996] explain that quinone and semi-quinone structures of humic acid becomes excited when absorbed UV radiation and react with molecular oxygen to yield superoxide ions. These ions can participate in the decomposition of some pollutants e.g. in a photochemical dechlorination of methoxychlor. The alkylperoxy radical RO<sub>2</sub><sup>•</sup> represents another type of radicals formed in aqueous solutions containing DOM during UV irradiation. As evaluated by Mill et al. [Mill T. 1980], RO<sub>2</sub><sup>•</sup> concentration at 10<sup>-9</sup> M in their investigations of cumene and pyridine photooxidation.

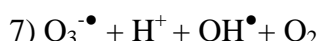
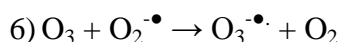
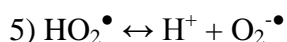
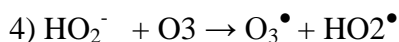
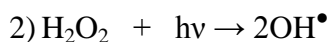
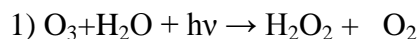
**Ozone (O<sub>3</sub>):** Ozone is an allotrope of oxygen that is much less stable than the diatomic allotrope (O<sub>2</sub>), breaking down with a half life of about half an hour in the lower atmosphere, to normal dioxygen. Ozone is sparingly soluble in water. At 20°C, the solubility of 100 percent ozone is only 570 mg.L<sup>-1</sup>. It is a powerful oxidant, second only to the hydroxyl free radical, among those chemicals typically used in water treatment. Therefore, it is capable of oxidizing many organic and inorganic compounds in water. It was found that the concentration of humic acids in most types of water decreases during the process of ozonization [Killops 1986] due to decomposition of these structures. The decomposition of humic acids follows through gradual decay to compounds with a simpler structure, till the resulting species have a molecular mass of less than 1000, then being easily biodegradable [Gilbert 1988]. Gilbert and Killops S.D [Killops 1986] examined indicated that 3-4 mg O<sub>3</sub> / per mg of DOC are enough to decrease the chemical oxygen demand by 60%. Ozone decomposes spontaneously during water treatment by a complex mechanism that involves the generation of hydroxyl free radicals. Under the influence of the short wavelength UV, Oxygen molecules react to form ozone.



But when irradiated with 200-300 nm light it decomposes rapidly in a chain reaction. The original mechanism for atmospheric ozone formation and destruction from oxygen species was suggested by Chapman [Chapman 1930].

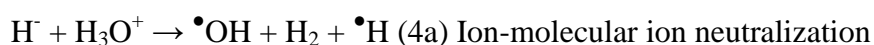
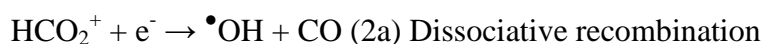
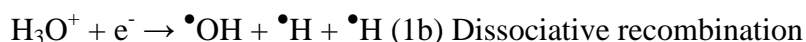
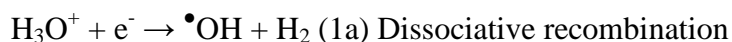


Where O<sub>2</sub><sup>\*</sup> represents excited oxygen molecules. Irradiation of Ozone with UV radiation in aqueous solution forms hydroxyl radicals (OH<sup>•</sup>), which are non-specific oxidants. Various pathways of OH<sup>•</sup> formation are possible [Chelkowska 1992]. Peroxy (HO<sup>2-</sup>) and superoxy (O<sup>2-</sup>) ions are being formed as intermediates in the dissociation of H<sub>2</sub>O<sub>2</sub>.



The oxidation rate always increased in oxidant/UV system as compared to systems containing Oxidants ( $\text{H}_2\text{O}_2$ ,  $\text{O}_3$  etc) only [Yue 1992].

**Hydroxyl radicals ( $\text{OH}^\bullet$ ) and hydrogen peroxide ( $\text{H}_2\text{O}_2$ ):** Hydroxyl radicals are highly reactive and consequently short life time forming an important part of radical chemistry. Most notably hydroxyl radicals are produced from the decomposition of hydroperoxides ( $\text{HOOH}$  or  $\text{ROOH}$ ) or, in atmospheric chemistry, by the reaction of excited atomic oxygen with water. Photooxidation under the influence of sunlight (Solar-UV) [Woldemichael 2011] have a radical character forming  $\text{OH}^\bullet$  radical in which their subsequent reaction with organic matter results a natural biodegradation [Mill 1989]. The hydroxyl radical can damage virtually all types of macromolecules such as carbohydrates, nucleic acids (mutations), lipids (lipid peroxidation) and amino acids [Reiter 1995]. Studies of  $\text{OH}^\bullet$  distribution [Harju 2000] suggest that in dense gas,  $\text{OH}^\bullet$  is mainly formed by dissociative recombination of  $\text{H}_3\text{O}^+$ . Dissociative recombination is the reaction in which a molecular ion recombines with an electron and dissociates into neutral fragments. Important formation mechanisms for  $\text{OH}^\bullet$  are:

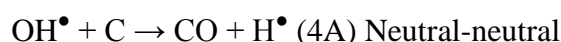
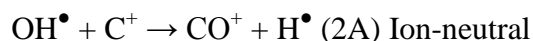
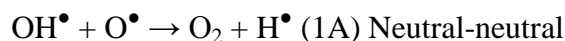


Experimental data on association reactions of  $\text{H}^\bullet$  and  $\text{OH}^\bullet$  suggest that radiative association involving atomic and diatomic neutral radicals may be considered as an effective mechanism

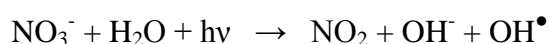
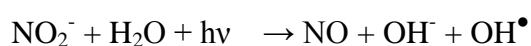


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for the production of small neutral molecules in the interstellar clouds [Field D. 1980]. We can see that atomic oxygen takes part both in the production and destruction of  $\bullet\text{OH}$  as shown below.

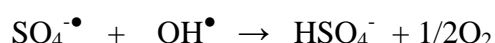
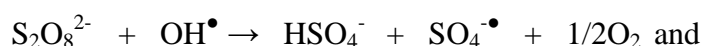


The influence of the addition of  $\text{H}_2\text{O}_2$  on the efficiency of UV-digestion of many organic compounds in aqueous solutions was studied [Yue 1992]. An often used decomposition method consists of acidifying (pH = 2) mineralized samples containing  $\text{H}_2\text{O}_2$  and subsequent photooxidation. An additional effect of acidifying is the dissociation of labile metal complexes of organic compounds. In the presence of nitrates and nitrites natural water can generate  $\text{OH}^\bullet$  radicals [Kotzias 1982].



**Persulfate ion ( $\text{S}_2\text{O}_8^{2-}$ ):** One of the strongest oxidizing agents in aqueous medium is the persulfate ion which shows a radical character at the first stage.  $\text{S}_2\text{O}_8^{2-} \rightarrow 2\text{SO}_4^{\bullet-}$

This reaction is initiated by sunlight or by substances contained in the aqueous solution. Obviously the reaction forms hydroxyl radical in the second stage by the reaction of sulfate ions with water.  $\text{SO}_4^{\bullet-} + \text{H}_2\text{O} \rightarrow \text{HSO}_4^- + \text{OH}^\bullet$  and  $\text{S}_2\text{O}_8^{2-}$  undergoes a chain reaction in the next steps as follows



The strong oxidizing properties of  $\text{S}_2\text{O}_8^{2-}$  ions, due to the formation of  $\text{H}_2\text{O}_2$ ,  $\text{O}_2$  and  $\text{OH}^\bullet$  as reaction products, means that persulfate is commonly used in mineralization by UV radiation [Van Steenderen 1981]. However the degree of mineralization varies depending on

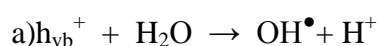
the type of the catalyst, their concentration, and intensity of UV radiation, temperature and the pH of the solution under examination.

**Titanium dioxide (TiO<sub>2</sub>):** In recent decades, the photocatalytic degradation of various toxic organic compounds (particularly organochlorides and nitrogenous compounds) has been proposed as a viable process to detoxify drinking water [Bahnemann 1994, Bahnemann (1999, Pichat 1997, Ollis 1993, Robert 2002]. UV irradiating pulverulent semi-conductors like TiO<sub>2</sub> in suspension or fixed to various supports in aqueous solutions containing organic pollutants, creates a redox environment able to destroy these pollutants. This property of TiO<sub>2</sub> is well demonstrated and documented in literature [Blake 1997]. Many researchers have shown that most organochloride compounds as well as many pesticides, herbicides, surfactants and colorings are completely oxidized into non-toxic products like carbon dioxide, hydrochloric acid and water. For instance, in 1998 EPA (US Environmental Protection Agency) made an inventory of more than 800 molecules that can be degraded by this process (Table 2.4).

**Table 2.4:** *Main ranges of pollutants*

Utilization	Products
Chlorinated solvents	Chloroform, carbon tetrachloride, trichloroethylene, chlorobenzene...
Non-chlorinated solvents	Acetone, acetonitrile, benzene, cyclohexane, formaldehyde, phenol, methylbenzene...
Insecticides	Aldrin, dichlorvos, lindane, parathion, monocrotophos...
Pesticides	Atrazine, monuron....
Dyes	Acid orange 7, green malachite, naphthol blue black....
Detergent	Octoxynol (triton X-100)...

The basic process of photocatalysis consists of ejecting an electron from the valence band (VB) to the conduction band (CB) of the TiO<sub>2</sub> semiconductor creating a 'h<sup>+</sup>' hole in the valence band ( $\text{TiO}_2 + h\nu \rightarrow e_{\text{cb}}^- + h_{\text{vb}}^+$ ). This is followed by the formation of extremely reactive radicals like OH<sup>•</sup> at the semi-conductor surface and/ or a direct oxidation of the organic species(R).



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This shows that the ejected electrons react with electron acceptors like oxygen adsorbed or dissolved in water or the electrons and holes may recombine together without electron donor or acceptor such as this.  $e_{cb}^- + h_{vb}^+ \rightarrow \text{TiO}_2$ .

The above process demonstrated that, many organic compounds could be photooxidized to CO, using  $\text{TiO}_2$  as a catalyst [Matthews 1987, Grlitzel 1987, Izumi 1980].

**Other oxidizing agents:** There are numbers of oxidants other than those discussed previously which are applied in photooxidation decomposition of organic substances like DOM. The application of  $\text{KMnO}_4$  and  $\text{K}_2\text{Cr}_2\text{O}_7$  in photooxidation was studied by Korolev et al. [Korolev 1983] and Frimmel and Winkler [Frimmel 1975],  $\text{Ce}(\text{SO}_4)_2$  by [Korolev 1983, Blazka 1983],  $\text{UO}_2(\text{N}_3)_2$ , and  $\text{OsO}_4$  by [Blazka 1983] and Pt by [Korolev 1983].

### c. Analytical Application of UV-photocatalysis

In general ultraviolet (UV) digestion is utilized mainly in the treatment of uncontaminated or slightly contaminated natural water matrices (aqueous solutions) such as sea water, surface water, fresh water, river water, lake water, ground water, estuaries, and coastal water. Solar photocatalytic mineralization of dissolved organic (DO) water pollutants using interaction between UV radiation with and without semi-conductor catalysts and or/ Oxidants has a strong potential in the industrial destruction of toxic organics in water or mineralization of DOM for different analytical procedures has been widely demonstrated in recent years [Woldemichael 2011, Goswami 1997b, Herrmann 1999, Malato 1999, Bockelman 2000] and the number of applications and target compounds are numerous. In zones with medium/high solar radiation solar detoxification is useful for treating water contaminants with maximum organic concentration of several hundreds of  $\text{mg.L}^{-1}$ , non-biodegradable contaminants and biocides and hazardous contaminants present within complex mixtures of organics. Methods of determination of elements in solution require homogenous samples. UV-digestion in general and Solar-UV decomposition in particular can be taken as a preliminary step in the analysis of heavy metals, carbon, nitrogen and phosphorus [Golimowski 1985]. The first use of UV photooxidation as a digestion process of water samples for subsequent determination of carbon, nitrogen and phosphorus was done in 1966 by Armstrong et al. [Armstrong 1966]. This method was applied as pretreatment step for determination of metals (mercury) in 1975 by Frimmel and Winkler [Frimmel 1975]. These reports originated the study on UV digestion, especially by electroanalysts, who determined the metal concentrations in waters by voltammetry. The UV-photooxidation became most important for water and waste water

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analysis, because of low concentration of investigated elements, so the application of other digestion methods could cause the contamination. Metals are determined in various types of matrices by means of different techniques, of which VA and AAS are the most important once. In the determination of almost all metals the mineralization process is usually conducted in acidic medium to avoid a precipitation except for chromium which is determined in neutral medium. Large number of studies deal with the determination of metals in UV digested natural waters. For instance cadmium, copper, lead and zinc [Golimowski 1990, Woldemichael 2011], lead and cadmium [Sipos 1979], mercury [Frimmel 1975], chromium [Golimowski, 1985], iron [Golimowski] 1975], arsenic [Stringer 1979], antimony and bismuth [Postupolski 1991) nickel, cobalt and Uranium [Woldemichael 2012] were determined. Optimal conditions of artificial UV mineralization were assessed by Labuda et al. [Labuda 1994]. The objective was to decompose high content of humic acid in water. They used UV irradiation to eliminate the negative effect of the presence of humic acid on Voltammetric determination. The method is also applied to digest organic matrices in waste water in which the concentration of Cu, Pb, Cd, Hg and Zn [Kolb 1992], Fe [Golimowski 1989] and arsenic [Stringer 1979]. Studies also proved that UV mineralization eliminates the observed non-homogeneity due to the metal participation in bacterial metabolic process. Similarly solar UV mineralization of DOM in river was carried out for voltammetric trace analysis of cadmium, cobalt, copper, lead, nickel, uranium and zinc [Woldemichael G. 2011, Woldemichael 2012]. In summary UV mineralization with and without oxidants has shown great application in electroanalytical techniques especially in the field of voltammetry.

## ***2.2. Voltammetric methods of trace metals determination***

Voltammetry is an electrochemical method in which current is measured as a function of the applied potential in a process of changing electrode potential, or the faradaic current or both with time. Historically, it was developed from the discovery of polarography in 1922 by the Czech chemist Jaroslav Heyrovsky (Nobel prize winner, 1922). The common characteristic of all voltammetric techniques is that they involve the application of a potential ( $E$ ) to an electrode and the monitoring of the resulting current ( $i$ ) flowing through the electrochemical cell. Voltammetric cells (instrumentation) consists of a working (indicator) electrode, a reference electrode, and usually a counter (auxiliary) electrode, stirrer and the gas line. The working electrode is where the reaction or transfer of interest is taking place. The reduction or oxidation of a substance at the surface of a working electrode, at the appropriate applied

potential, results in the mass transport of new material to the electrode surface and the generation of a current. Generally, working electrodes are made of platinum, gold, mercury, and carbon. Mercury is the classical electrode for polarography which is termed as dropping mercury electrode (DME). This is the only electrode where the surface of the electrode is renewed with each new drop. Hanging mercury drop electrode (HMDE) and static mercury drop electrode (SMDE) are commonly used in voltammetry. To avoid toxic mercury, many solid electrodes are introduced and are becoming popular. In particular, electrodes made of carbon such as glassy carbon, graphite, carbon paste, and carbon fibers have gained popularity. In addition to these, Mercury, gold, bismuth, and other metals can be deposited as thin metal films on carbon and serves as thin metal film electrodes (TMFE) with excellent analytical advantages in trace metal analysis. The selection of working electrode is determined by the redox potential of the analyte and the potential window within which the solvent and the supporting electrolyte remains electrochemically inert. Silver–silver chloride (SSC), and saturated calomel electrode (SCE) are the most common reference electrodes. The counter electrode is the current carrying electrode and it must be inert and larger in dimension. Platinum wire or foil is the most common counter electrode (Table 2.5). Even though voltammetric techniques may appear to be very different at first glance, their fundamental principles and applications derive from the same electrochemical theory.

**Table 2.5: Electrode materials and properties**

Type of electrode	Materials	Electrolyte	Properties
Working Electrode(W)	Platinum	1M H <sub>2</sub> SO <sub>4</sub>	-0.25 to 1.2V
	Platinum	1M KOH	-1.0 to 0.6V
	Mercury	1M H <sub>2</sub> SO <sub>4</sub>	-1.2 to 0.5V
	Mercury	1M KCl, NaOH	-2.0 to 2.0V
	Carbon(Glassy carbon, carbon paste, graphite, diamond film)	1M HClO <sub>4</sub> , 0.1M KCl	-2.0 to 1.5V -1.5 to 1.0V
Reference electrode (R)	Ag/AgCl, satd. KCl (SSC)	0.197 V vs. NHE	At 25°C NHE- Normal hydrogen electrode
	Hg (l)/Hg <sub>2</sub> Cl <sub>2</sub> (s), satd. KCl (SCE)	0.241 V vs. NHE	
Counter electrode (C) or Auxiliary electrode	Platinum, carbon or steel	Similar to the working electrode except for steel	Must be larger than W

Voltammetric techniques have large analytical advantages such as excellent sensitivity with a very large useful linear concentration range for both inorganic and organic species ( $10^{-12}$  to

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10-11 M)[Fifield 2000, Barek 2001b], a large number of useful solvents and electrolytes, a wide range of temperatures, rapid analysis times (seconds), simultaneous determination of several analytes, the ability to determine kinetic and mechanistic parameters, a well developed theory and thus the ability to reasonably estimate the values of unknown parameters[Sures 2007] and the ease with which different potential waveforms can be generated and small currents measured[Zhang 2002]. A wide linear range, portable and low cost instrumentation, speciation capability and a wide range of electrode allows analysis of many types of samples in many fields of application. As an indication, it has been applied in the analysis of environmental samples [Buffle 2005], pharmaceutical samples [Hilali 2003, Carapuc 2005], food samples [Volkoiv 2001, Karadjova 2000], dye samples and forensic samples [Woolever 2001], DNA-Analysis at heated carbon paste electrodes [Wang 2000]. DNA Hybridization detection [Flehsig 2005], Adsorptive stripping voltammetric detection of daunomycin at mercury and bismuth alloy electrodes [Duwensee 2007] and Heavy metals in natural waters [Woldemichael 2011, Woldemichael 2012]. Voltammetry can be performed with a nano-electrode for the detection of single molecular events [Schuster 1998]. Similar electrodes can be used to monitor the activity of neurotransmitter in a single living cell in sub-nanoliter volume electrochemical cell and measurement of fast electron transfer kinetics [Bratten 1997]. Trace metal analysis with electrically heated Bismuth-Film electrode for Voltammetric Stripping Measurements of Trace Metals was made by Flehsig et al [Flehsig 2002]. The introduction of modern electronics, computers, softwares and new materials enhanced voltammetric techniques to be applied in hundreds of new analysis in several fields as described above. Voltammetric sensors appear to be an ideal candidate for miniaturization and mass production. This is evident in the development of lab-on-chip [Ahuja 2006] technologies with applications ranging from capillary electrophoresis chips with integrated electrochemical detection [Wooley 1998], chemical plume tracking [Kikas 2002], micro-electrophoresis system for explosive analysis [Hilmi 2000], and clinical diagnostic devices [Eggers 1995, Selvaganapathy 2001, Lorraine 2003]. Continuous improvement and diversification of voltammetric techniques enabled the analysis of hundreds of chemicals and electroactive species. Some of these techniques are:

ii) Direct Voltammetry which includes linear sweep voltammetry (LSV), direct current voltammetry (DCV), and Hydrodynamic voltammetry (HDV).

ii) Pulse methods such as Normal pulse voltammetry (NPV), Differential pulse Voltammetry (DPV) Square wave voltammetry (SWV) and Cyclic voltammetry (CV).

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iii) Stripping methods like Anodic stripping voltammetry (ASV), Cathodic stripping voltammetry (CSV), Adsorptive stripping voltammetry (AdSV) and Adsorptive catalytic stripping voltammetry (AdCSV).

Though the basic principles behind all techniques are the same, all of them are employed for the determination of specific categories of organic and inorganic analytes as discussed earlier.

### ***2.3. Ecotoxicological aspects of some heavy metals***

Heavy Metals mainly include the transition metals, some metalloids, lanthanides, and actinides. The definitions for heavy metals have been given in different ways based on density, atomic number, atomic weight, chemical properties or even toxicity and ecotoxicity [Duffus 2002]. In similar document, sometimes the term is loosely used to refer to both the element and its compounds based on categorization by density, which is rarely a biologically significant property. Definitions given based on density lacks consistency. For instance, heavy metals are those with elemental densities above 7 g.cm<sup>3</sup> [Bjerrum 1936], greater than 4 g.cm<sup>3</sup> [Grant 1987], and greater than 5 g.cm<sup>3</sup> [Parker 1989]. Heavy metals are also defined based on their atomic weights [Lewis 1993], Atomic Number [Hale 1988]. Other defined heavy metals based on chemical properties and Toxicity [Birchon 1965, Merriman 1965, Hampel 1976, Scott 1981]. The later is most applied to this literature and the research itself. In this work the term heavy metals refers to those metals / Metalloids having chemical properties which are toxic or ecotoxic at trace levels( in µg.L<sup>-1</sup> level or below) to humans, animals and plants.

#### **2.3.1. Heavy metal contamination of water and the resulting health hazards**

Heavy metals, enter the environment and / or water from many sources, including industrial effluents, automobile exhaust, mining operations, pesticides, urban run-off, atmospheric deposition agricultural activity and even natural soil [Bately 1996, Haynes 2000]. Heavy metal pollutants are conservative and most of them are essentially permanent additions to the environment often being highly toxic to biota if their concentration is above certain threshold limit [Clark 1992]. Unnatural quantities of metals such as arsenic (As), cadmium (Cd), copper (Cu), mercury (Hg), lead (Pb), nickel (Ni) and zinc (Zn) have been released, and continue to be released into the aquatic environment through storm water and waste- water discharge. As, Cd, Cu, Hg and Zn are the five metals with most potential impact that enter the environment in elevated concentrations as a consequence of agricultural activities. Potential impacts from heavy metals are generally restricted to locations adjacent to major cities or industrialized areas on the coastal fringe or river banks [Haynes 2000] and to site draining areas of intensive

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agriculture. Once introduced into the aquatic environment, heavy metals have a potential to affect sediment nutrient cycling, cell growth and regeneration, as well as reproductive cycles and photosynthetic potential of aquatic organisms [Peters 1997]. Heavy metal contamination of soil results from anthropogenic activities such as mining [Galas-Gorchev] 1991). Also it comes from smelting procedures [Clemente 2008], agricultural practices [Stimpfl 2006] as well as natural activities. Chemical and metallurgical industries, plastic and rubber industries, various consumer products and from burning of waste containing these products are the most important sources of heavy metals in the environment and eventually end up in aquatic system via leaching and wash down of the soil and the air. Heavy metals get accumulated in time in soils and plants and could have a negative influence on physiological activities of plants (e.g. photosynthesis, gaseous exchange, and nutrient absorption), determining the reductions in plant growth, dry matter accumulation and yield. In small concentrations, the traces of the heavy metals in plants or animals are not toxic and some of them (e.g. Co and Zn) are even vital for life. [Lewis 1993, de-Vries 2007], cadmium and mercury are exceptions; they are toxic even in very low concentrations [Razo 2004]. Monitoring the endangerment of soil with heavy metals is of interest due to their influence on groundwater and surface water and also on plants, animals and humans [Nam 2005]. The health hazards and adverse effects resulting from acute and / or chronic exposure to heavy metals are complicated and extremely dangerous in which some of the effects are irreversibly harmful. An exposure to significantly higher cadmium levels is known to produce toxic effects on humans and animals. It accumulates in kidneys, where it damages filtering mechanisms. This results the excretion of essential proteins and sugars from the body and further kidney damage. According to the Agency for toxic substances and disease registry (ATSDR), acute or chronic exposure to cadmium causes diarrhea, stomach pain, severe vomiting, bone fracture, reproductive failure or even infertility, damage to central nervous system and immune system and psychological system. In similar document, acute uptake of cobalt ends up in vomiting, and nausea, vision and heart problems and thyroid damage. Many heavy metals are carcinogenic, immunogenic or mutagenic and very low concentration. They can affect cardiovascular system, reproductive and nervous systems [Lars 2003, US-EPA 1998]. Heavy metals such as zinc (Zn), copper (Cu), chromium (Cr), arsenic (As), cadmium (Cd), and lead (Pb) are potential bioaccumulative toxins of the dairy production system [Li 2005]. The young are more prone to the toxic effects of these heavy metals, as the rapidly developing body systems in the fetus, infants and young children are far more sensitive. ATSDR states that, childhood exposure to some metals can result in learning difficulties, memory impairment, damage to the nervous



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system, and behavioral problems such as aggressiveness and hyperactivity. At higher doses, heavy metals can cause irreversible brain damage. Children may receive higher doses of metals from food than adults, since they consume more food for their body weight than adults. The extent of harmfulness of exposure to heavy metals or heavy metal compounds depend on factors such as the dose (how much), the duration (how long), and how one come in contact with it. We must also consider any other chemicals you are exposed to and your age, sex, diet, family traits, lifestyle, and state of health.

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## 3. Methods and Materials

### 3.1. Instrumentation and Operating Conditions

The instrument we used in both the DpASV and DpAdSV experiment is a  $\mu$ Autolab potentiostat (Ecochemie) with General Purpose Electrochemical System (GPES) 4.9 software package. The electrolysis cell was a Metrohm 663VA stand. The set up is applied in the determination of both the concentrations and pseudopolarography of heavy metals such as cadmium, cobalt, copper, lead, nickel, uranium and zinc. A three electrode system is applied with a HMDE as working electrode, a glassy carbon counter electrode, and Ag/AgCl (3 mol.L<sup>-1</sup> KCl) reference electrode. ASV method was used for the measurement of both the concentration, pseudopolarography and recovery rates of zinc, cadmium, lead and copper in Warnow and Awash River water samples While AdSV was used for the determination of the concentration, recovery test and pseudopolarography of nickel, cobalt and uranium in the different samples of the above two river waters. The specific instrumental operating conditions for both ASV and AdSV are summarized in table 3.1

**Table 3.1:** Instrumental operating parameters for DpASV and DPAdSV analysis of Zn (II), Cd (II), Pb (II), Cu (II), Ni (II), Co (II) and U (VI) in river water samples

Instrumental operating parameters	Analytes		
	Zn(II), Cd(II), Pb(II), Cu(II)	Ni(II), Co(II)	U(VI)
Method	DpASV	DpAdSV	DpAdSV
Working electrode	HMDE	HMDE	HMDE
Calibration	Standard addition	Standard addition	Standard addition
Purging time (s)	10 to 600	10 to 600	10 to 600
Deposition potential (V)	-1.15	-0.7	0.1
Deposition time (s)	120	120	30
Equilibration time (s)	10	5	5
Modulation amplitude(V)	0.0202	0.0202	0.05
Modulation time (s)	0.05	0.05	0.05
Interval time (s)	0.5	0.10	0.10

Start potential (V)	-1.15	-0.8	0.04
End potential (V)	0.05	1.2	-0.21
Voltage step(V)	0.049	0.049	0.006
Scan rate (V/s)	0.01	0.013s	0.012

The UV-Vis spectra of the original river water samples and the artificial water containing humic acid was determined by Cary 100 Bio Spectrophotometer operated by Cary series winUV software.

### ***3.2. Reagents***

We obtained all reagents from certified manufacturers (Merck, www.merk.de, Fluka, www.sigmaaldrich.com) as they are ultrapure grades.  $\text{Cd}^{2+}$ ,  $\text{Co}^{2+}$ ,  $\text{Cu}^{2+}$ ,  $\text{Ni}^{2+}$ ,  $\text{Pb}^{2+}$ ,  $\text{U}^{6+}$  and  $\text{Zn}^{2+}$  ( $1000 \text{ mg.L}^{-1}$ ) stock solutions were diluted with ultrapure water (SG water, www.water-siemens.com;  $\rho > 18.2 \text{ M}\Omega\text{.cm}$ ,  $\text{TOC} < 2 \text{ }\mu\text{g.L}^{-1}$ ). Also we used similar grade nitric acid (65%) to acidify the water sample at the spot of sampling. This is done to avoid adsorption of the metal ions to the walls of the containers and to hinder biological activities. The supporting oxidizing agent was  $\text{H}_2\text{O}_2$  (30%) in its purest grade. The excess hydrogen peroxide was removed by addition of amidosulfonic acid and hydroxysulfonic acids prior to the determination. Humic acid (20% ash content) was used to prepare “artificial river water” containing approximate concentration of the acid as in natural river water. We prepared two kinds of buffer solutions for the determination of the metals in ASV and AdSV methods. Sodium acetate buffer with 0.1M KCl (pH = 4.6) was used as supporting electrolyte in ASV determination of Zn(II), Cd(II), Pb(II) and Cu(II) While Ammonia ( $\text{NH}_3/\text{NH}_4\text{Cl}$ ) buffer (pH= 9.5) was applied in AdSV measurement of Ni(II) and Co(II). This supporting electrolyte was prepared from ammonia solution ( $w_{\text{NH}_3} = 25\%$ ) and hydrochloric acid ( $w_{\text{HCl}} = 30\%$ ). The ligands that we have used in AdSV of nickel and cobalt were  $0.1 \text{ mol.L}^{-1}$  of dimethylglyoxim (DMG) in ethanol solution and triethanolamine (TEA) in water (1:1). In the determination of uranium, freshly prepared chloranilic acid (CAA) solution was applied as a complexing agent according to the earlier method [Sander 1994].

### ***3.3. Overview of the sampling sites***

Warnow River Water samples were collected from the downstream of Warnow River (Rostock, Germany) at sampling site of  $54^\circ 05' 38.12'' \text{N}$ ,  $12^\circ 09' 06.33'' \text{E}$  and used in the study of the application of solar UV photooxidation as sample pre-treatment step in ASV and AdSV methods of trace metals analysis. Awash River water samples were collected from

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Mid-Upper Awash River (Ethiopia) at sampling sites labeled as Koka dam, Sire Robi, Wonji Kuruftu, Wonji Drain and Awash Malkasa (table 4.9). These samples are used for practical application of Solar UV photooxidation in natural water sample pre-treatment step in voltammetric trace metals determination. Awash River originates from central plateau of Ethiopia near *Ginch-West shewa*. Flowing south east direction to the plains of south west Shewa, pass to the southern side of Addis joining tributaries like Akaki which originates from Addis and a collection of many streams and Municipal waste drains and Modjo River which joins Awash near Koka Dam. Each sampling site has its own unique ecological and geological characteristics. Koka Dam sampling site is located at the upstream of the dam from the incoming river. The upstream of this site is characterized by low velocity, high turbidity and more shallow nature. Any pollution detected at this point can be considered as a cumulative effect of the pollution due to Akaki River (a collection of small streams of Addis Ababa) which is highly saturated with urban runoff and industrial wastes of the city, Akaki industrial zone waste disposal sites, landfills and agrochemicals leached from Ada'a and the surrounding regions. Sire Robi is located at Adama drinking water treatment plant just below Koka Dam. The sample is taken from the water that is coming out from power station generating hydroelectric power at Koka hydroelectric power station. The velocity of the water is relatively high because of the drop of the altitude (43 m) across short distance (3 km). Awash River water joins Modjo River in Koka Lake. This river is the most deadly polluted river, (nearly waste water) because of the disposal of industrial wastes like Modjo tannery, Awash tannery, Ethiopian Tannery, Addis Modjo oil mills etc (field observation).



**Figure 3.1:** Map of Awash River sampling sites (Map adopted from Google map 2012)

There are also large horticultural fields located near by the lake irrigating the horticultures with Modjo River or Lake Water. Therefore the sample taken from sire Robi helps to

determine the cumulative effect of pollutants from all the above point and non point sources. Wonji Kuruftu is located just at the start point of the river diversion to irrigate Wonji sugarcane plantation. Downstream of Sire Robi to Wonji Kuruftu there is no significant industrial and agricultural activity taking place. The next sample was collected from within the plantation, from the drained Awash river to irrigate the sugarcane plantation and this sample is labeled as Wonji drain. This sample does not represent the river water, but helps to analyze the effect of the plantation on the environment in general and the ecosystem in particular. The last sample is collected from Awash Malkasa, a site located at the end of Wonji sugarcane plantation, where it is expected to include all the leached agrochemicals from the plantation, industrial wastes from pulp and paper, Wonji and Wonji Shewa sugar plant and Wonji candies and sweet factory.

**Table 3.2:** *Awash River sampling sites-Ethiopia*

Sampling site	GPS coordinates	Altitude (m)
Koka Dam	8°24`24.02`N, 039°01`11.66`E	1595
Sire Robi	8°28`45.82`N, 039°10`52.41`E	1553
Wonji Kuruftu	8°28`24.37`N, 039°12`47.98`E	1553
Wonji Drain	8°23`39.38`N, 039°19`16.11`E	1545
Awash Malkasa	8°24`04.91`N, 039°20`03.28`E	1542

### ***3.4. Sample collection, pretreatment and preservation***

Sample collection protocol is the most important step to be followed in any analysis of environmental samples such as water, air or soil. In this experiment we followed USEPA Guidelines for regulatory monitoring and testing water and waste water protocol. Accordingly, new plastic bottles were labeled and transported to the sampling site. The bottles were rinsed twice with sample water before they are filled with water sample. All the samples were grab samples taken from the surface at one point in the cross-section of the flow. Following the protocol, the samples are then acidified with ultra pure HNO<sub>3</sub>, to pH of 2±0.2 (1mL of 65% HNO<sub>3</sub> per liter of the sample), packed and transported to the laboratory for further preparation. In the laboratory, they were filtered through with a 0.45 μm pore size cellulose acetate membrane filter inserted in a Millipore filtration glass assembly and, then, 100 μmol.L<sup>-1</sup> of ultrapure H<sub>2</sub>O<sub>2</sub>, was added and the same aliquot of Warnow River water samples were then divided into three sample holders designated as Original Sample, SoUV Sample, and UV Sample. The Original Sample was that part of the sample with no UV treatment and stored in refrigerator at 4°C until determination. The Original sample served as

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control sample throughout the experiment. The SoUV Sample was transferred to a UVA transparent (330–450 nm) polyethylene terephthalate (PET) bottles and exposed to a solar radiation of UVA intensity of  $3.6\text{mW}/\text{cm}^2$  for 6 and 12 hours as needed. We transferred the UV Sample to a UV-transparent (200 - 450 nm) quartz glass tube. This was irradiated for 6 and 12 hours by means of a UV lamp (30 W and 254 nm). Determination of Zn(II), Cd(II), Pb(II) and Cu(II) was done with the 6 hours SoUV and UV irradiated samples while the determination of Co(II), Ni(II) and U(VI) was done for 6 and 12 hours SoUV irradiated and 6(12) hours UV irradiated sample. The control sample (Original sample) was analyzed for all seven heavy metals under the same experimental conditions with the irradiated samples. Usually the solar irradiation time was between 10:00AM and 4:00 PM because this duration is the ideal time where maximum efficiency of solar radiation is achieved as a result of increase of the incident angle of the radiation. A parabolic aluminum solar collector is used to increase the intensity of solar radiation which also increases the infrared intensity of the incident radiation. The synergetic effect of elevated water temperature and UV radiation enhances the digestion process [McLaughlin 2004]. All the three samples were stored in a refrigerator at  $4^\circ\text{C}$  until determination. We prepared artificial water containing  $1.0\text{mg}\cdot\text{L}^{-1}$  and  $0.1\text{mg}\cdot\text{L}^{-1}$  of humic acid in order to compare and estimate the concentration of humic acid in the original Warnow river water by means of UV-Vis spectrophotometer. The filtered, acidified and  $\text{H}_2\text{O}_2$  added Awash River water samples were irradiated with solar UV-Radiation of intensity  $5.4\text{mW}/\text{cm}^2$  for about 12 and 24 hours and shipped to Rostock-Germany for metals determination. In Rostock Laboratory, portion of the 24 hours SoUV sample was taken and irradiated with a 30W (254 nm) artificial UV radiation for 6 hours and determined for its trace metals' concentration.

### ***3.5. Application of the methods to trace metals analysis in River water samples***

The determination of all the seven metals in all samples was carried out by taking 20 mL of the samples with the corresponding buffer and supporting electrolytes and complexing agents. In the case of the determination of cadmium, copper, lead and zinc, 2.0 mL of  $0.1\text{mol}\cdot\text{L}^{-1}$  of the pH of the cell was adjusted to 4.6 by adding sodium acetate buffer containing  $0.1\text{mol}\cdot\text{L}^{-1}$  KCl. This serves to enhance electrical conductivity of the solution too. Similarly ammonia buffer was added as a supporting electrolyte and also to adjust the pH of the solution to 9.5 while  $150\mu\text{L}$  of  $0.10\text{mol}\cdot\text{L}^{-1}$  of DMG in ethanol and  $150.0\mu\text{L}$  of triethanolamine (TEA) in a

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1:1 water solution were added as a complexing agent in the determination of nickel and cobalt. For uranium we added 500  $\mu\text{L}$  of 5  $\text{mmol.L}^{-1}$  (= 0.12  $\text{mmol.L}^{-1}$  in the cell) CAA as complexing agent. The pH of this cell was tuned to  $2\pm 0.18$  with  $\text{HNO}_3/\text{NaOH}$ . All samples aerated by purging with nitrogen for 600s, at initial and for 10s in between the successive measurements. The four heavy metals (cadmium, copper, lead and zinc) were analyzed simultaneously by ASV. Nickel and cobalt were analyzed by AdSV simultaneously while determination of uranium was carried out in similar method (AdSV) separately as described in section 3.1. In all determinations standard addition method is used for calibration. Pseudopolarographic data was recorded with the same equipment for the same aliquot of Original, SoUV and UV samples with varying accumulation potentials. Accumulation potential ( $E_{\text{acc}}$ ) was scanned between  $-1.20\text{ V}$  and  $-0.60\text{ V}$  for Zinc, and  $-1.2\text{ V}$  and  $0.10\text{ V}$  for copper with scan increment ( $E_{\text{sc.Inc}}$ ) of  $0.05\text{ V}$  in both cases. Potential scan range for nickel was  $-1.5\text{ V}$  to  $0.20\text{ V}$  with scan increment of  $0.05\text{ V}$ . Similarly pseudopolarograms of uranium was recorded in between  $-0.15\text{ V}$  and  $-0.05\text{ V}$  with scan increment of  $0.01\text{ V}$ . To estimate the concentration of humic acid in the original river water sample, UV-VIS spectra was taken for 1000 and 100-fold diluted original Warnow river water sample and compared with artificially prepared river water containing  $1\text{ mg.L}^{-1}$  and  $0.1\text{ mg.L}^{-1}$  humic acid.

### ***3.6. Determination of Method detection limit (MDL)***

EPA defines the method detection limit (MDL) as, "the minimum concentration of a substance that can be measured and reported with 99% confidence that the analyte concentration is greater than zero and is determined from analysis of a sample in a given matrix containing the analyte. It is a statistical concept, based on the ability of a measurement method to determine an analyte in a sample matrix, regardless of its source of origin. It should always be above the instrumental detection limit (IDL). Instrument Detection Limit (IDL) is the concentration equivalent to a signal, due to the analyte of interest, which is the smallest signal that can be distinguished from background noise by a particular instrument. According to the above method, the procedure required seven replicates of samples spiked at the approximate concentration up to 5 times the manufacturers instrumental detection limit (IDL). The calculated MDL was evaluated using several checks to determine if it does or does not meet all of the necessary criteria. Accordingly, the spiked level is below 10 times the observed MDL but greater than MDL itself for all analytes.

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## 4. Results and discussion

### *4.1. Application of Solar UV- Photooxidation in the determination of trace metals in River water samples by Anodic stripping Voltammetry.*

The main aim of this study is to evaluate the effect of Solar UV photooxidation on the decomposition (digestion of) dissolved organic matter in river water samples. We investigated its application in River Water or natural water samples pre-treatment (digestion) step in Stripping Voltammetric trace metals' analysis. The experiment was carried out based on the instrumental and experimental conditions given in sections 3.1 to 3.3. The concentration of Zn (II), Cd (II), Pb (II) and Cu (II) was measured by DpASV and Voltammograms were obtained for Original, SoUV and UV samples (Fig. 4.1a-c). The standard addition method is used to plot calibration curves (fig. 4.2 to 4.5) and we calculated the concentrations of the metals in all of the three samples (Original, SoUV and UV) from these curves. The corresponding numerical values obtained from calibration curves are given in Table 4.1. Accordingly, the concentrations of Zn(II) was found to be  $17 \pm 0.43 \mu\text{g.L}^{-1}$  in the original undigested sample,  $22.86 \pm 0.41 \mu\text{g.L}^{-1}$  in 6 hours Solar UV irradiated sample and  $26 \pm 0.40 \mu\text{g.L}^{-1}$  in 6 hours artificial UV irradiated sample.

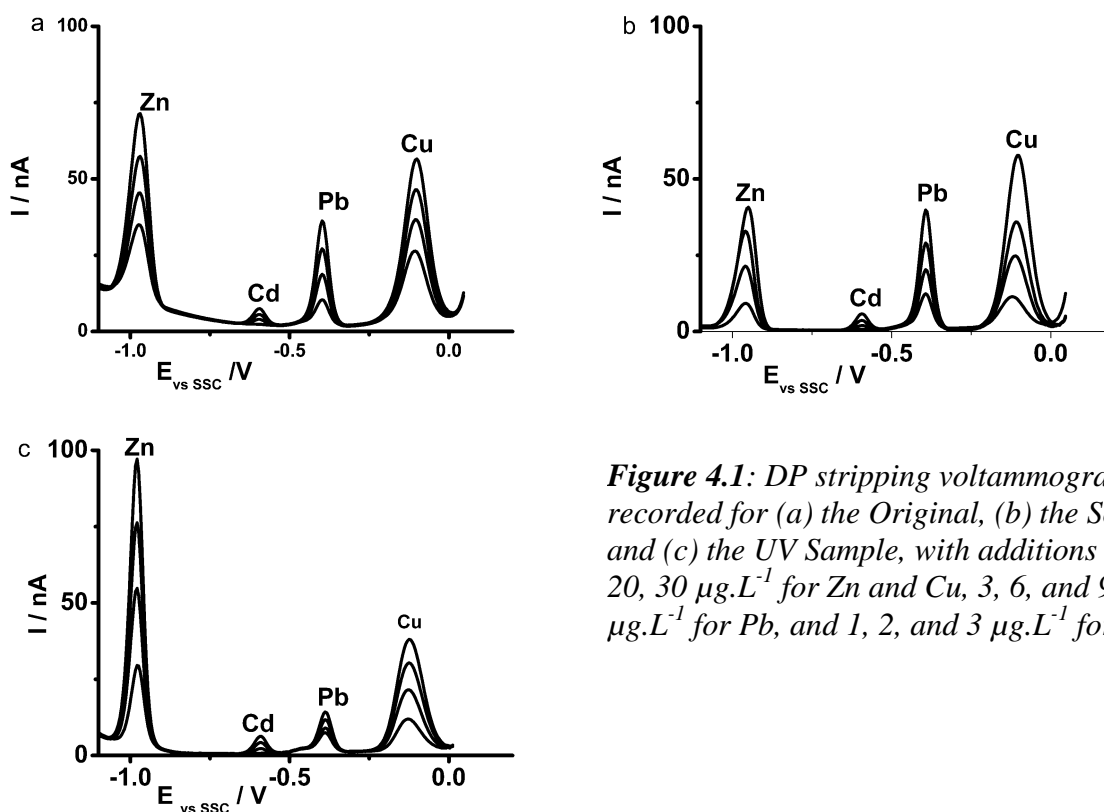
**Table 4.1:** Concentration and recovery rates measured for Zn, Cd, Pb, and Cu in Original Sample, SoUV Sample and UV Sample of Warnow River

Element	Concentration of samples ( $\mu\text{g.L}^{-1}$ )			Recovery rate of samples (%)		
	Original	SoUV	UV	Original	SoUV	UV
Zinc	$17.17 \pm 0.43$	$22.86 \pm 0.41$	$26.67 \pm 0.40$	$88.81 \pm 0.20$	$95.91 \pm 0.13$	$92.90 \pm 0.21$
Cadmium	$0.03 \pm 0.01$	$0.05 \pm 0.03$	$0.05 \pm 0.005$	$75.32 \pm 0.15$	$87.11 \pm 0.22$	$93.66 \pm 0.11$
Lead	$4.36 \pm 0.05$	$7.07 \pm 0.02$	$7.50 \pm 0.01$	$84.21 \pm 0.32$	$90.32 \pm 0.10$	$94.14 \pm 0.31$
Copper	$32.20 \pm 0.34$	$58.85 \pm 0.31$	$73.57 \pm 0.28$	$133.59 \pm 0.42$	$112.75 \pm 0.37$	$104.16 \pm 0.35$

The concentration of Cd (II) was calculated from calibration curves in similar method and the numerical values are  $3 \pm 0.00$ ,  $0.05 \pm 0.00$ , and  $0.05 \pm 0.01 \mu\text{g.L}^{-1}$  for Original, SoUV and UV samples respectively. Pb (II) also showed increase in concentration with both solar and



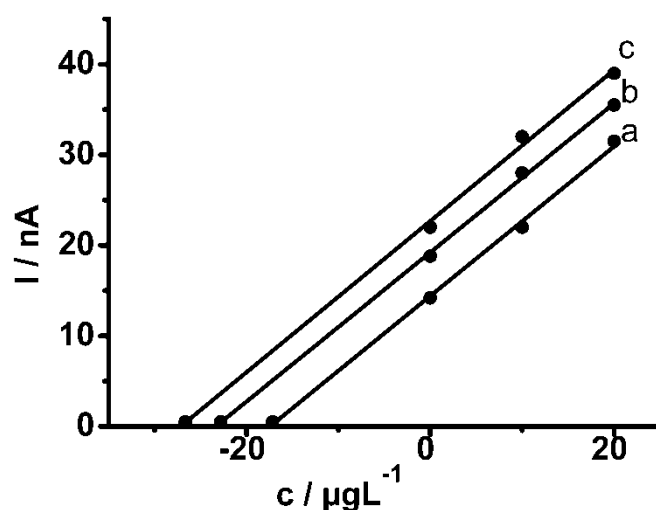
artificial UV irradiation. This improvement in the concentration is given as  $4.36 \pm 0.05$ ,  $7.07 \pm 0.02$ , and  $7.50 \pm 0.01 \mu\text{g.L}^{-1}$  for Original, SoUV and UV samples respectively. In the case of Cu (II), concentration was found to be  $32.20 \pm 0.34 \mu\text{g.L}^{-1}$  for original,  $58.85 \pm 0.31 \mu\text{g.L}^{-1}$  for SoUV, and  $71.57 \pm 0.28 \mu\text{g.L}^{-1}$  for UV samples. From this experiment we observed that the concentration of each of the tested four metals increased with both solar and artificial UV irradiation.



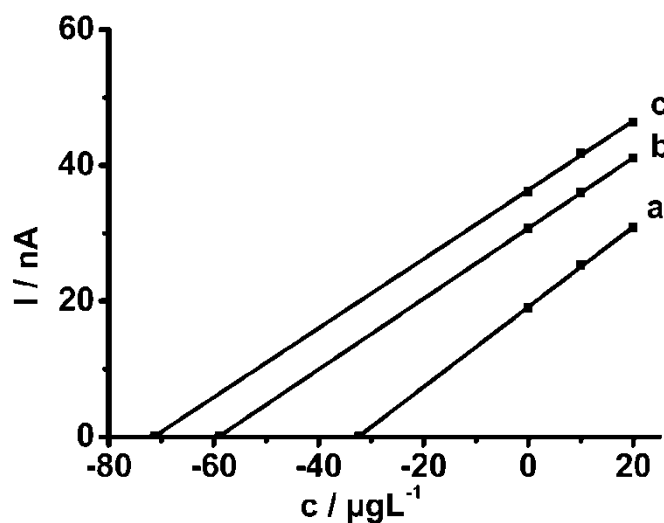
**Figure 4.1:** DP stripping voltammograms recorded for (a) the Original, (b) the SoUV and (c) the UV Sample, with additions of 10, 20, 30  $\mu\text{g.L}^{-1}$  for Zn and Cu, 3, 6, and 9  $\mu\text{g.L}^{-1}$  for Pb, and 1, 2, and 3  $\mu\text{g.L}^{-1}$  for Cd.

The effect of UV irradiation is different for different metals. In this case UV irradiation is more effective in the case of Pb (II). The effect of SoUV is almost same as artificial UV. In the case of Zn (II) and Cu (II), we have observed improvement of the amount of free metal ions available for voltammetric determination. However 6 hours of Solar UV irradiation seems to be insufficient to complete the digestion. From these findings we observed that most of the matrix components are digested. However, more than 6 hours of solar UV irradiation is needed to complete the decomposition at the given latitude. Cd (II) was least affected by the irradiation process which seems that Cd (II) is to be less prone to complexation by components present in this river water matrix. We used the recovery experiment to evaluate the data obtained by means of stripping voltammetry in the various sample treatment procedures taken into consideration. To calculate the recovery rates, a known concentration of

the metal ions were added to all the three categories of the samples prior to the determination. The calculated recovered concentrations and percentage recovery (recovery rates) are tabulated in table 4.1. From this experiment we observed that the Original sample showed the worst recovery rates confirming that more DOM is present complexing considerable amount of the metal ions. Some metals yielded best recovery rate with solar irradiation. This can be seen for Zn (II) (fig.4.2). The recovery rates of all the three samples of Cu (II) exceeded 100%. This can be related to the observed decrease in slope of the standard addition function (Fig. 4.3). In this case the sensitivity in presence of organic river water matrix is slightly higher compared to both radiation-treated samples due to complexing matrix component which leads to increased stripping signal. This phenomena leads to incorrect analysis result though the absolute stripping signals are larger for the irradiated samples

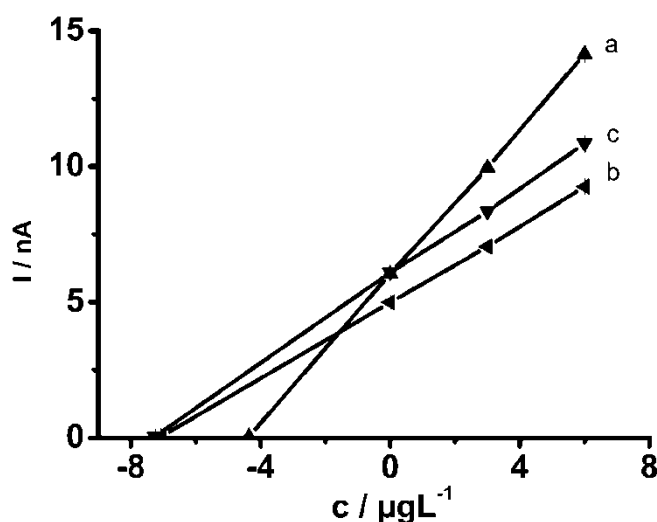


**Figure 4.2:** Standard addition for Zn (II) in (a) Original, (b) SoUV and (c) UV Samples.

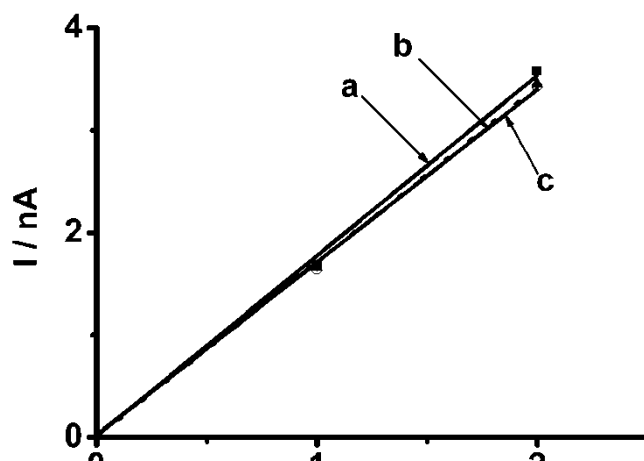


**Figure 4.3:** Standard addition for Cu (II) in (a) Original, (b) SoUV, and (c) UV Sample.

The Original sample of Cu (II) showed unusual recovery rate (133.6%) which can be associated to considerable complexing agent. Pseudopolarographic studies also confirmed the above fact. Pseudopolarographic measurements were used to study the nature of matrix-complexed Zn (II) and Cu (II) ions in all the three samples labeled as Original, SoUV and UV as presented in Fig. 4.6 and 4.7. The significant increase in peak currents is mainly the result of irradiation treatment.

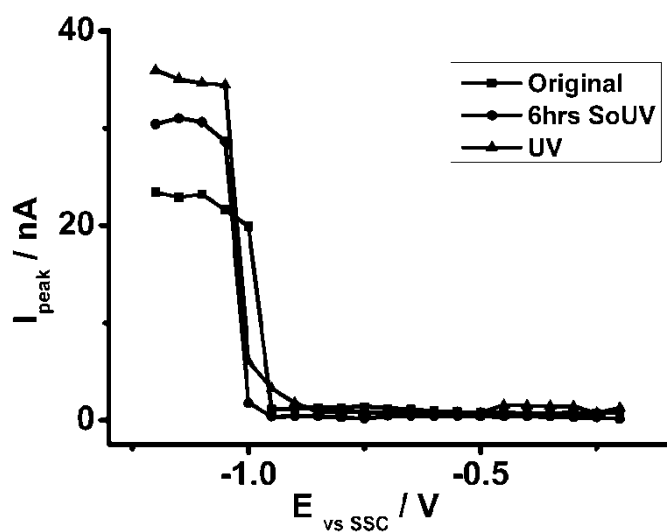


**Figure 4.4:** Standard addition calibration curve for lead in (a) Original, (b) SoUV and (c) UV Samples.



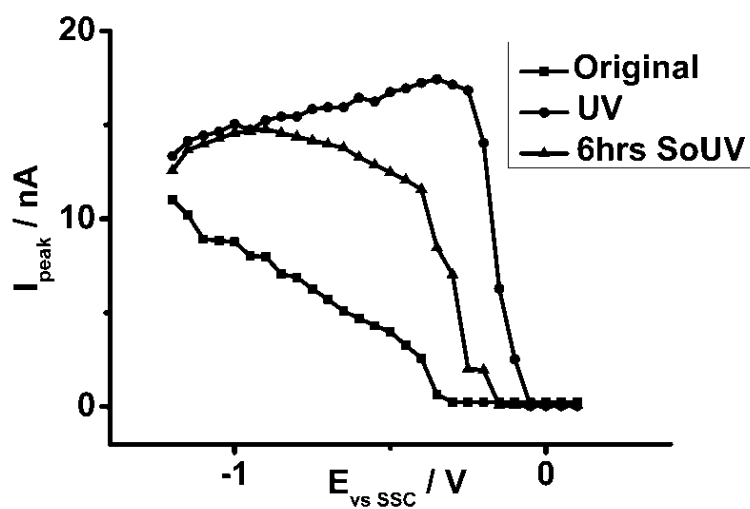
**Figure 4.5:** Standard addition calibration curve for Cd (II) in (a) Original, (b) SoUV and (c) UV Samples.

This would mean that, more metal ions are available for reduction during the deposition step after artificial UV or solar UV irradiation. The results also confirmed that the irradiative digestion treatment is more complete for artificial UV light. We also proved that solar irradiation results a remarkable release of Zn (II) ions from the matrix complexes by destruction of the organic ligands. Fig. 7 illustrates the Dramatic effect of both irradiation treatment methods tested in Pseudopolarographic study of Cu (II). We observed the change of both the Pseudopolarographic half-wave potential and the step height in considerable extent after treatment with either solar or artificial UV light.

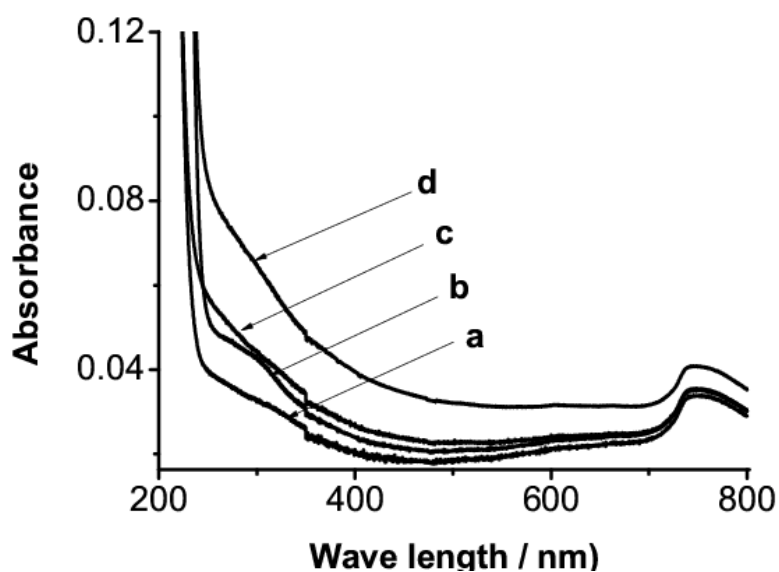


**Figure 4.6:** Pseudopolarograms of  $10 \mu\text{g.L}^{-1}$  zinc in (a) Original Sample, (b) SoUV Sample, and (c) UV Sample; DPASV at HMDE, accumulation time 240 s, initial potential  $-1.2 \text{ V}$ , final potential  $-0.70 \text{ V}$ , scan increment  $0.05 \text{ V}$ , interval time  $0.5 \text{ s}$ , scan rate  $10 \text{ mV/s}$ , and  $0.1 \text{ M}$  acetate buffer containing  $0.1 \text{ M}$  KCl ( $\text{pH} = 4.6$ ).

Fig. 4.8 represents UV-Vis spectra of 1000- and 100 fold diluted Warnow river water sample and  $1 \text{ mg.L}^{-1}$  and  $0.1 \text{ mg.L}^{-1}$  of an artificially prepared humic acid solution. From this figure it is possible to estimate the concentration of humic acid in the original river water sample. Thus the concentration of the most abundant UV/Vis active substance, humic acid is estimated to be as much as  $1000 \mu\text{g.L}^{-1}$ . The irradiation treatment effect can be explained in respect to the high level of complexing organic matter in the Original sample.



**Figure 4.7:** Pseudopolarograms of  $10 \mu\text{g.L}^{-1}$  copper in (a) Original Sample, (b) SoUV Sample, and (c) UV Sample; DPASV at HMDE, accumulation time 240 s, initial potential  $-1.2 \text{ V}$ , final potential  $0.10\text{V}$ , scan increment  $0.05\text{mV}$ , interval time  $0.5 \text{ s}$ , scan rate  $10 \text{ mV/s}$ , and  $0.1\text{M}$  acetate buffer with  $0.1\text{M}$  KCl ( $\text{pH} = 4.5$ ).

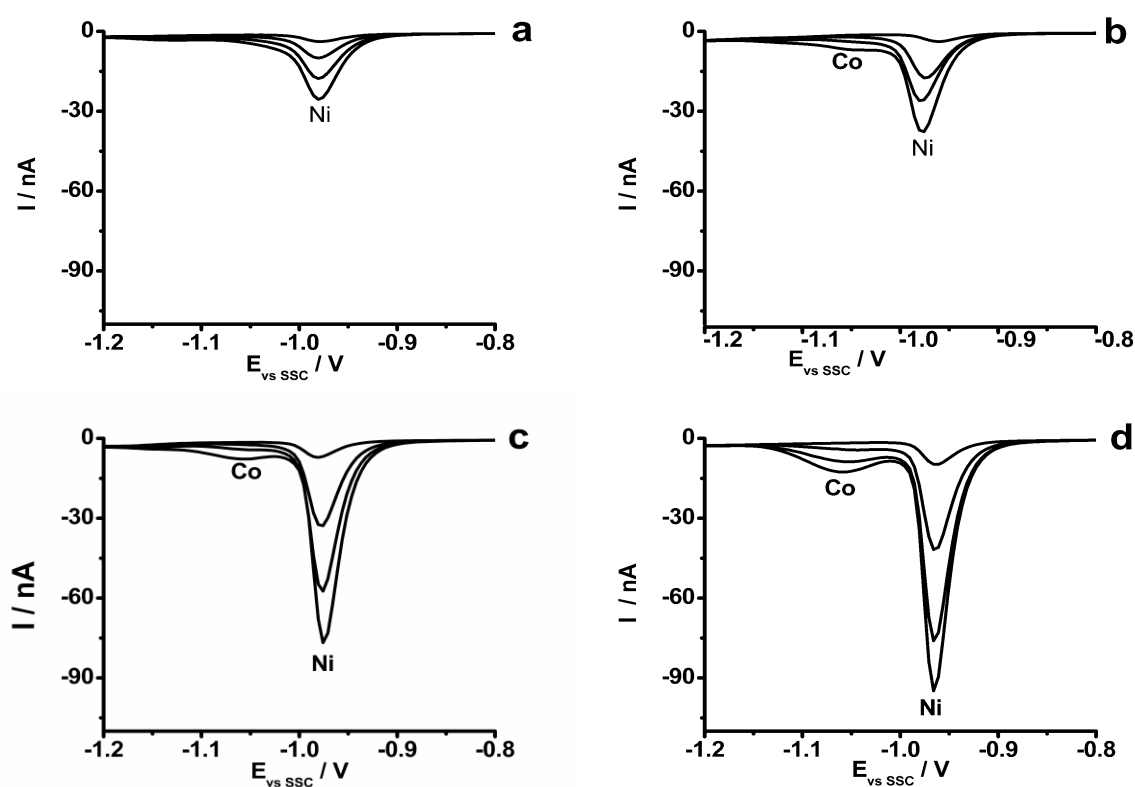


**Figure 4.8:** UV-VIS spectra of (a)  $0.1\text{mg.L}^{-1}$  humic acid, (b)  $1 \text{ mg.L}^{-1}$  humic acid, (c) Warnow river sample 1: 1000 diluted, and (d) Warnow River.

## 4.2. Solar UV-assisted pre-treatment of river water samples for the voltammetric determination of nickel and cobalt ultra-traces

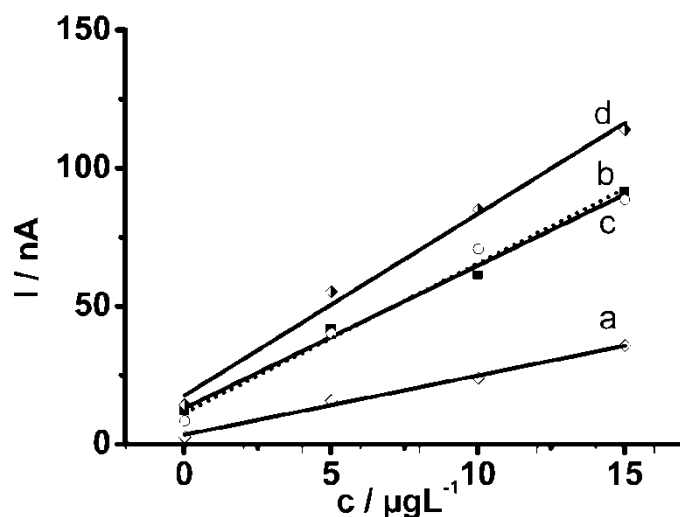
### 4.2.1. Standard addition experiments and recovery rates

In AdSV determination of Ni (II) and Co (II), we used DMG-TEA as complexing agent. However, these ligands compete with the natural ligands (e.g. humic acid) that are already present in the river water samples. The presence of the natural ligands makes the metal ions unavailable for stripping analysis. Therefore, it is the most important step to destroy the natural matrices using appropriate sample pre-treatment method. In our case this has been accomplished by solar UV photooxidation method. In other words, we applied the previously investigated Solar UV-assisted river water sample pre-treatment method [Woldemichael 2011].



**Figure 4.9:** Voltammograms of nickel(II) and cobalt (II) for a) Original Sample, b) 6 hours SoUV Sample, c) 12 hours SoUV Sample, and d) UV Sample measured in the presence of ammonia buffer ( $\text{NH}_3/\text{NH}_4\text{Cl}$ ,  $\text{pH} = 9.5$ ) as a supporting electrolyte and DMG-TEA as complexing agents. The standard concentrations were  $5, 10, 15 \mu\text{gL}^{-1}$  for nickel (II) and  $1, 2, 3 \mu\text{gL}^{-1}$  for cobalt (II)

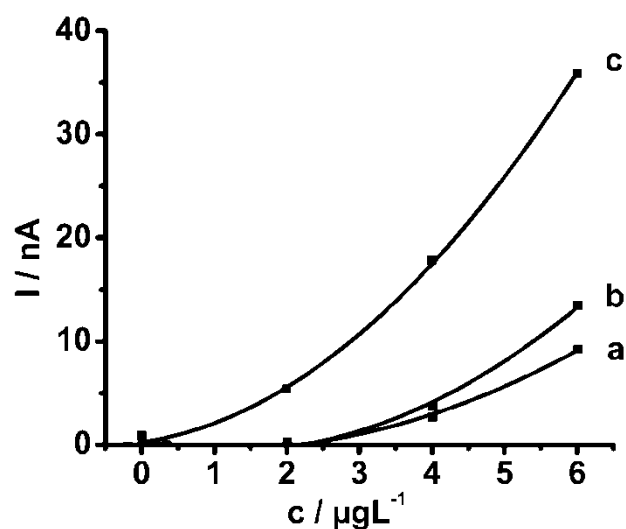
The method is tested for its effectiveness in AdSV determination of Ni (II) and Co (II) ultra-traces. The samples were collected, prepared and determined as indicated in section 3 of this dissertation. DpAdSV method is applied in the determination of the concentration, Recovery rates and the Pseudopolarographic analysis. Fig. 4.9 represents voltammograms of Ni (II) and Co(II) in river water samples pretreated with UV radiation of different source and time of exposure. We used standard addition method for calibration and used to calculate the concentrations. The calibration curves corresponding to the two metals are given in figures 4.10 and 4.11. The calculated concentration values for Ni (II) and Co (II) are given in table 4.2. These values demonstrated that, the concentration of Ni (II) increased by 28% up on solar UV irradiation for 6 hours. Extended irradiation for another 6 hours has increased the amount of free metal ion by 58%. This value is very close to the 63% value obtained for the sample irradiated with a 30 W artificial UV lamp for 6 hours. In the case of Co (II), the original sample showed no or little increase in peak current even after the addition of  $8 \mu\text{g.L}^{-1}$  of Co (II) standard solution. This is a proof that the amount of DOM in Warnow river sample is capable of complexing all cobalt ions present in the sample plus the amount added from outside. Similarly, no significant peak current is detected after irradiating the samples with solar UV for 6 and 12 hours.



**Figure 4.10:** Standard addition calibration curves for nickel(II) in a) Original Sample, b) 6 hours SoUV Sample, c) 12 hours SoUV Sample, and d) 6 hours UV Sample. Added standard concentrations of nickel were 5 and 10 and 15  $\mu\text{g.L}^{-1}$ .



These samples also absorbed up to  $2 \mu\text{g.L}^{-1}$  of Co (II) ions before a significant peak current appears. This indicates complexing organic matter was still present in the water consuming the above amount of the metal ions. Thus extending time of irradiation beyond 12 hours or increasing the intensity of solar UV radiation is needed to complete the digestion. The observed significant increase in cobalt peak current for artificial UV irradiated sample confirms the above fact. In the later case, we applied square function for regression fit and to calculate the concentration of Co (II). Accordingly  $0.33\mu\text{g.L}^{-1}$  of Co (II) was detected in this sample. The overall experimental findings suggest that 12 hours of solar UV irradiation under the given UV intensity could not complete the digestion process and hence considerable amount of the metal ions are bound by the undigested ligands. This is also the main reason for the observed deviation of the cobalt calibration curve from linearity. Absorption of cobalt ions by the complexing DOM leads to the application of Co (II) standard solution for titration of the complexing compounds in the river water sample. In general, this experiment confirmed that 12 hours of solar UV irradiation cannot complete the digestion process under the above experimental conditions (Central Europe and  $3.6\text{mW.cm}^{-2}$ ). This would imply that, more intense solar UV radiation such as equatorial or tropical sun is needed to complete the digestion in Co (II) determination by the recommended method.



**Figure 4.11:** Standard addition calibration curves for Co(II) in a) a) 12 hours SoUV Sample, b) 6 hours SoUV Sample, and c) 6 hours UV Sample. Added concentrations of Co were 2, 4 and  $6 \mu\text{g.L}^{-1}$ .

We confirmed the effect of UV irradiation from the improvement of the recovery rates (tables 4.2 and 4.3). In the case of Co (II), the concentration was calculated indirectly from the percentage of DOM that had been decomposed by solar and artificial UV irradiation which intern is obtained from the

recovery experiments. This approach has solved the difficulty of calculating Co (II) concentration from calibration curves due to substantial deviation of the curves from linearity especially for the 6 and 12 hours solar UV irradiated samples. The recovery rate of Ni (II) has been determined by the usual and conventional method. We spiked  $5 \mu\text{g.L}^{-1}$  of Ni (II) standard solution (concentration to be recovered) to the samples at the beginning and determined the percentage recovery by standard addition method (table 4.2). The effect of the UV irradiation has been confirmed from successive increment of the percentage recovery. The end result of the recovery rate implies that nearly all complexing organic ligands are destroyed as a result of UV irradiation of the samples

**Table 4.2:** Concentration of Nickel in the Original Sample, 6 and 12 hours SoUV irradiated and 6 hours artificial UV irradiated UV Sample.

	<i>Original</i>	<i>SoUV</i>		<i>UV</i>
	<b>0hrs</b>	<b>6hrs</b>	<b>12hrs</b>	<b>6hrs</b>
Concentration ( $\mu\text{g.L}^{-1}$ )	$1.66 \pm 0.06$	$2.12 \pm 0.10$	$2.62 \pm 0.06$	$2.71 \pm 0.02$
Recovery Rate (%)	$87.80 \pm 0.95$	$90.80 \pm 0.55$	$95.20 \pm 0.43$	$99.00 \pm 0.42$
Blank recovery rate (%)		$100.48 \pm 0.20$		

In the case of Co (II), only  $3.60 \mu\text{g.L}^{-1}$  (45.00 %) of the spiked  $8 \mu\text{g.L}^{-1}$  was recovered in the original sample. This shows that about  $4.40 \mu\text{g.L}^{-1}$  or (55%) of the spiked Co (II) was absorbed by the DOM existing in the river water sample. The concentrations of the standard addition in the case of Original sample were 8 and  $16 \mu\text{g.L}^{-1}$  Co (II). The amount of cobalt ions complexed in the irradiated samples were much smaller compared to the Original samples. We spiked only  $2 \mu\text{g.L}^{-1}$  of the Co (II) standard solution of which 65, 72 and 86.50 percent were recovered after 6 hours SoUV, 12 hours SoUV and 6 hours artificial UV irradiation. The recovery tests clearly showed that, too much DOM is present in the Original Sample. As has been demonstrated above the natural complexing organic matter in this sample is enough to complex  $4.40 \mu\text{g.L}^{-1}$  of the spiked Co (II) and the unknown concentration of the originally complexed Co (II). The complexing capacity of the ligands in the river sample dramatically reduced to 0.59, 0.48 and  $0.27 \mu\text{g.L}^{-1}$  after 6 hours SoUV, 12 hours SoUV and 6 hours artificial UV irradiation. These values also indicate the equivalent concentration of the undigested complexing natural ligands after radiation treatment. This would mean that only 13.41, 10.90 and 6.14 percent of the original DOM was remaining after 6 and 12 hours solar and artificial UV irradiation, respectively. Therefore, 86.59, 89.10 and 93.86 percent of

the original complexing ligands were destroyed after 6 hours solar UV, 12 hours solar UV and artificial UV irradiation, respectively. Our experiment showed that, this digestion can be accomplished by solar UV irradiation. The only requirement is optimizing time of solar UV irradiation at locations of intense sun light such as tropical regions.

**Table 4.3:** Concentration of Cobalt in the Original Sample, 6 and 12 hours SoUV irradiated and 6 hours artificial UV irradiated UV Sample.

	Original	SoUV		UV
	0hrs	6hrs	12hrs	6hrs
Concentration ( $\mu\text{g.L}^{-1}$ )	0.00	0.00	0.00	0.33 + 0.02
Recovery Rate (%)	45.00 + 0.24	65.00 + 0.14	72.00 + 0.08	86.50 + 17
Blank recovery rate (%)		102.34 + 0.43		

**Table 4.4:** Recovery tests for Nickel. The given  $\text{Ni}^{2+}$  concentration values before and after spiking were determined by standard addition.

Nickel	Concentration ( $\mu\text{g.L}^{-1}$ )				Recovery rate (%)
	Before spiking	Spiked	After spiking	Recovered	
Original	1.66 ± 0.11	5.00 ± 0.01	6.05 ± 0.02	4.39 ± 0.12	87.80 ± 0.01
6 h SoUV	2.12 ± 0.01	5.00 ± 0.11	6.66 ± 0.10	4.54 ± 0.11	90.80 ± 0.02
12 h SoUV	2.62 ± 0.10	5.00 ± 0.12	7.38 ± 0.13	4.76 ± 0.23	95.20 ± 0.01
UV	2.71 ± 0.03	5.00 ± 0.01	7.660 ± 0.12	4.95 ± 0.02	99.00 ± 0.01
Blank	0.00 ± 0.03	5.00 ± 0.04	5.02 ± 0.03	5.02 ± 0.02	100.48 ± 0.01

#### 4.2.2. Pseudopolarography

The nature of freely available Ni (II) in the different samples was evaluated by means of Pseudopolarography. This experiment was not done for Co (II), because its level in all samples was too low to be detected by the method. Pseudopolarograms records of Ni (II) are given in fig.4.12. The results demonstrated a dramatic effect of the UV irradiation upon the stripping response of Ni (II) in the range between -1.05 and 0 V depositions potential. Whereas the maximal achievable voltammetric signals increased 10-fold for artificial UV and 7-fold for solar UV irradiation, also the potentials shifted towards positive direction. Both the Pseudopolarographic “half wave” potential and the current maximum potential were shifted by max. 300mV. This means that, with increasing UV irradiation dose, it becomes easier to form and adsorb the  $[\text{Ni}(\text{dmgH})_2]$  complex during the accumulation step in AdSV measurements. The effects of solar UV irradiation treatment were larger in case of nickel (II) and cobalt (II) compared with our earlier studies of zinc, cadmium, lead and copper

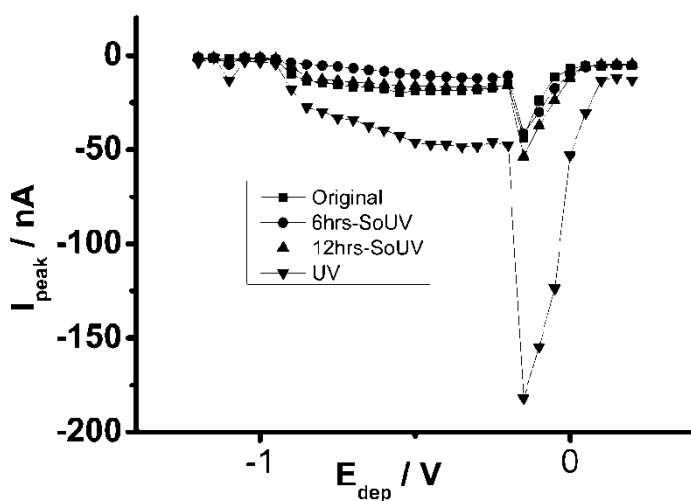
[Woldemichael G. 2011]. The main reason can be found in complexation of the analyte cations, which is on one hand needed for adsorptive accumulation. On the other hand, this desired process competes with complexation of the metal ions and humic substance ligands. The latter interactions seem to be strong as well. Another reason can be found in interfacial activity of the dissolved organic compounds in river water samples as they disturb the adsorptive accumulation of the analyte ion complexes with DMG.

**Table 4.5:** Recovery tests for cobalt. The given  $Co^{2+}$  concentration values before and after spiking were determined by standard addition.

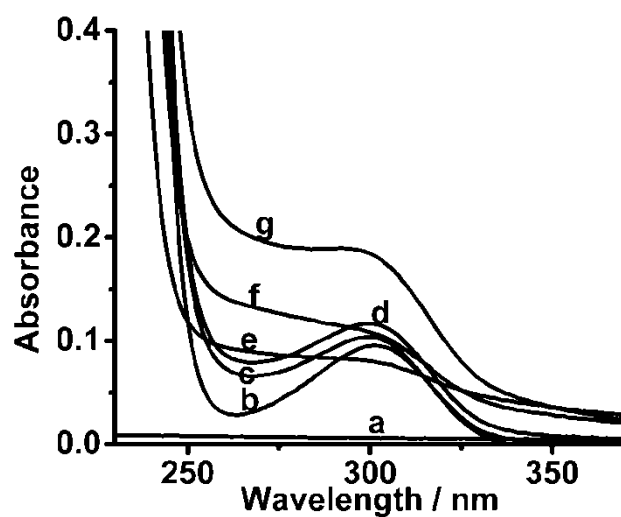
Cobalt	Concentration ( $\mu\text{g.L}^{-1}$ )			Recovered	Recovery rate (%)
	Before spiking	Spiked	After spiking		
Original	0.00	8.00	3.60	3.60	45.00
6hrs SoUV	0.00	2.00	1.30	1.30	65.00
12hrs SoUV	0.00	2.00	1.45	1.45	72.50
UV	0.33	2.00	2.06	1.73	86.50
Blank	0.00	2.00	2.05	2.05	102.34

#### 4.2.3. UV-Vis spectrophotometry

This experiment is designed to estimate the concentration of humic substances in the Original Warnow river water samples at the time of sampling. It is also used to estimate the extent of distraction of these substances as a result of radiation treatment base on the methods we applied in our previous work [Woldemichael 2011, Woldemichael 2012]. It is clear that the Major structure constituent of humic substances like humic acid, fulvic acid is aromatic ring. The observed UV/Vis Spectral absorption of the samples at 300 nm (fig. 4.13) confirms the above truth because this spectrum is a characteristic band of excitation of  $\pi$ -electrons in the benzene ring. The relative concentration of these humic substances can be deduced from the peak height of the spectra. We compare the concentrations bases on the spectral patterns. Thus the concentration of the artificial UV irradiated sample is less than  $10\text{mg.L}^{-1}$  while that of 6 hours Solar UV- irradiated sample is nearly  $10\text{mg.L}^{-1}$ . Similar evaluation reveals that the amount of dissolved organic matter (DOM) is decreased with intensified UV irradiation or by increasing radiation dose. This experiment is also used to estimate the concentration of humic substance in the original river water sample. As a result we can estimate the concentration of the humic substances in the Original river water sample to be 10-fold of  $10\text{ mg.L}^{-1}$  humic acid. In other words the river contains about  $100\text{ mg.L}^{-1}$  of humic acid.



**Figure 4.12:** AdSV pseudopolarograms of nickel measured at potential scan range of a) -1.5V to 0.20V and b) -1.2V to -0.2V in the presence of  $\text{NH}_3/\text{NH}_4\text{Cl}$  buffer at pH 9.5 as supporting electrolyte and DMG-TEA as complexing agents while other instrumental operating parameters are kept constant as given in table 1.

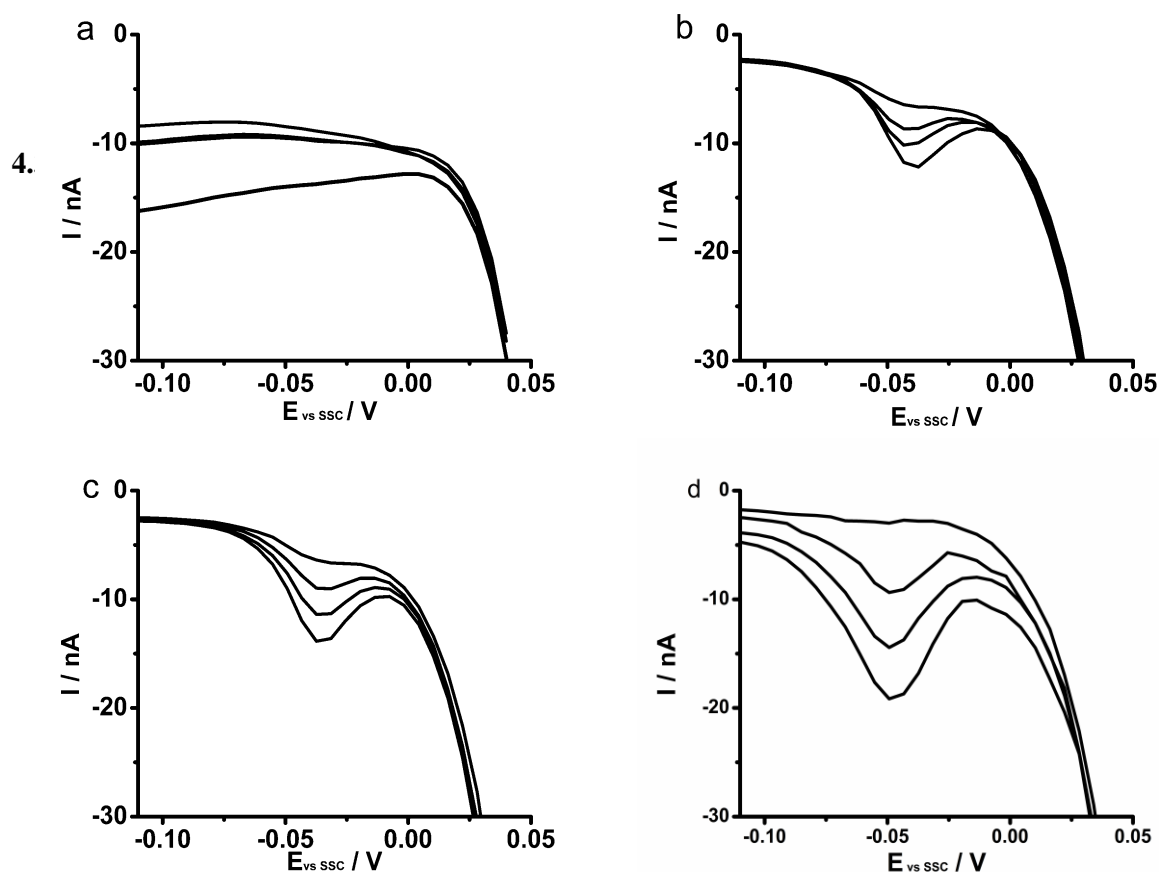


**Figure 4.13:** UV-Vis spectra for a) blank b) 6 hours UV Sample, c) 12 hours SoUV Sample, d) 6 hours SoUV Sample, e) 10 ppm humic acid, f) 2-fold diluted Original Sample, and g) Original Sample.

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### ***4.3. Solar UV-assisted sample preparation of river water for ultra-trace determination of uranium by adsorptive stripping voltammetry***

Uranium is one of the heavy metals existing in the environment in four oxidation states (III, IV, V and VI, the hexavalent being the most stable form). The ability of uranium to form various complex compounds with a quite large number of ligands makes it difficult to be measured with AdSV method because stripping analysis requires freely available metal ions. To accomplish this, we applied the cheap, green and efficient solar UV sample pre-treatment method as it has been done for zinc, cadmium, lead, cobalt and nickel in our earlier work [Woldemichael 2011]. We recorded voltammograms of different Warnow river water samples labeled as the Original untreated sample (Original), 6 hours solar UV treated sample (6hrs SoUV), 12 hours solar UV irradiated (12hrs SoUV) and 6 and 12 hours artificial UV treated sample (UV) (fig 4.14). The standard additions in the above determination are 2, 4 and 6  $\mu\text{g}\cdot\text{L}^{-1}$  of U (VI) standard solution in the presence of chloranilic acid (CAA) which served as complexing agent in Adsorptive stripping voltammetric determination of U (VI). Usually the peak will be observed for adsorptively accumulated uranium (VI) as a U-CAA complex on the surface of the HMDE at optimized CAA concentration ( $120 \mu\text{mol}\cdot\text{L}^{-1}$ ) and pH  $2\pm 0.12$ . The plot of the calibration curve (fig. 4.15) is employed to calculate the concentration of U (VI) in all samples. We tabulated the results obtained from the extrapolation of the standard addition calibration curve in table 4.6. In this experiment no U (VI) peak current was detected in the Original sample even after the standard additions (fig.4.14a). However, the UV radiation treated samples yielded typical peak current as seen from the voltammograms (fig.4.14b-d). It has been observed that the concentrations are increased greatly after UV irradiation. These values are  $0.88\pm 0.02$  for 6 hours SoUV sample,  $1.03\pm 0.11$  for 12 hours SoUV sample,  $1.07\pm 0.11$  for 6 hours artificial UV sample and  $1.12\pm 0.01 \mu\text{g}\cdot\text{L}^{-1}$  for 12 hours artificial UV samples. From these results we can conclude that, the method is effective in river water sample pre-treatment step. We can clearly see that, only 6 hours of solar UV irradiation could increase the concentration of freely available uranium by 88% even at Central Europe.

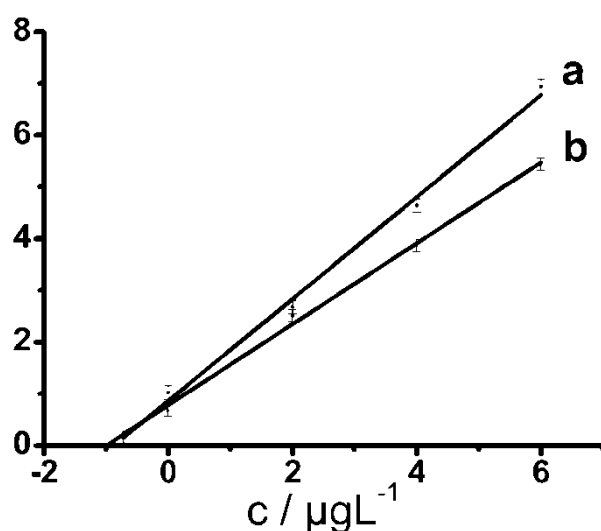


**Figure 4.14:** AdSV response of uranium(VI) in a) Original Sample b) 6 h SoUV, c) 12 h SoUV and d) 6 h UV Sample with standard additions of 2, 4 and 6  $\mu\text{g}\cdot\text{L}^{-1}$ . Potentials are given vs./AgCl (3 M KCl). AdSV conditions as in Table 1.

### 3.4.1. Recovery Rate

The determination of recovery rates for each category of samples confirmed the effect of UV radiation treatment on the amount of detectable U (VI) or the extent of decomposition of complexing organic matter in the samples. As we can read from table 4.7, nothing was recovered in the original sample. All the added uranium ions ( $2 \mu\text{g}\cdot\text{L}^{-1}$ ) were absorbed by the natural ligands present in the river water sample. Similar evaluation of the recovery rate showed that  $93.50 \pm 0.03 \%$  of the 6 hours SoUV Sample,  $94.00 \pm 0.02 \%$  of the 12 hours SoUV Sample,  $94.00 \pm 0.11$  and  $95.00 \pm 0.10 \%$  of 6 and 12 hours artificial UV Samples were recovered. These results are additional evidences for the effectiveness of our sample pre-treatment method. The observed blank recovery rate ( $108.00 \pm 0.02$ ) is in agreement with the expected contamination of the blank due to the cell itself and the reagents (CAA and  $\text{HNO}_3$ ) used for the determination as complexing agent and pH adjustment. From these results we confirmed that the solar UV irradiation effectively removed the complexing activity of DOM (Fig. 4.14a) in the river water samples there by releasing free U (VI) for voltammetric

detection (fig. 4.14b-d). As we have explained in previous paragraphs, even 6 hours of soft solar UV irradiation has a dramatic effect upon release of the U (VI) making it available for complex formation with CAA, and thus, AdSV analysis (Fig. 4.14b).



**Figure 4.15:** Standard addition calibration curves for uranium (VI) in a) SoUV Sample (6 hours) and b) UV sample (6 hours).

Irradiation with more intense and higher energy UV source (30 W, 254 nm) does not show much difference on the recovery rate though the sensitivity is increased (Fig. 4.14d). This is in contrast to our earlier findings for anodic stripping Voltammetry of Zn, Cd, Pb, and Cu [Woldemichael 2011], and also to AdSV analysis of Nickel and Cobalt in river water where artificial UV irradiation yielded the best recovery rates.

**Table 4.6:** The concentrations and recovery rates of uranium (VI) obtained in the Original Sample, 6 and 12 hour SoUV Samples and 6 and 12 hour UV Samples.

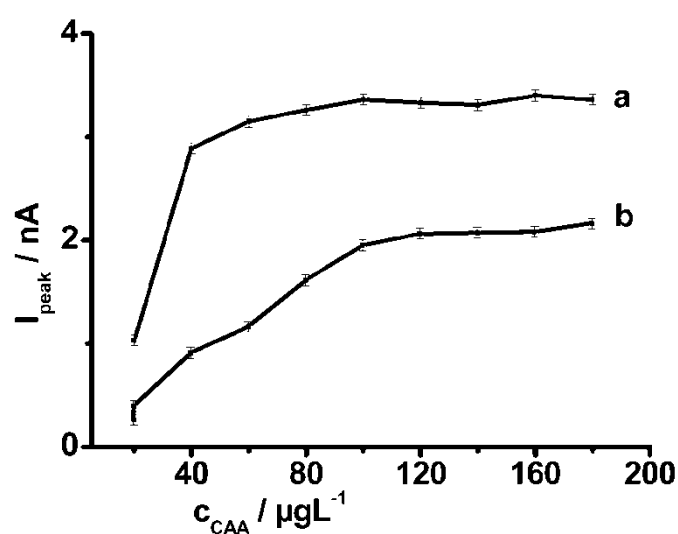
	Original	SoUV		UV		Blank
	0 hours	6 hours	12 hours	6 hours	12 hours	
Conc.( $\mu\text{g}\cdot\text{L}^{-1}$ )	0.00	0.88 + 0.02	1.03+ 0.02	1.07 + 0.11	1.12+0.01	0.00
Recovery (%)	0.00	93.50+ 0.02	94.00+0.02	94.00 + 0.11	95.00+0.01	108.00

#### 4.3.2. Effect of Ligand Concentration

We determined the optimum CAA concentration needed to complex the U (VI) in AdSV analysis. We compared the CAA concentration needed by the sample with a blank to which  $2 \mu\text{g}\cdot\text{L}^{-1}$  U (VI) was spiked. Accordingly the concentration of CAA required in the river water samples was much higher compared with the spiked blank sample. The spiked blank revealed that, almost no significant increase of peak current was observed beyond 60



$\mu\text{mol}\cdot\text{L}^{-1}$  CAA concentrations (Fig. 4.16a). To contrary, the peak current in the SoUV Sample was increasing up to  $120 \mu\text{mol}\cdot\text{L}^{-1}$  CAA concentrations (Fig. 4.15b). The large concentration of CAA needed by the sample could be associated with the competition between various complexing ligands present in the sample and the CAA itself. Therefore, we found that the optimum CAA concentration in the voltammetric cell should be  $120 \mu\text{mol}\cdot\text{L}^{-1}$  and we used this amount of CAA for all UV-irradiated samples. For original sample it was impossible to observe any peak at any CAA concentration even after the addition of  $100 \mu\text{g}\cdot\text{L}^{-1}$  U(VI) due to higher concentration of DOM in the sample.

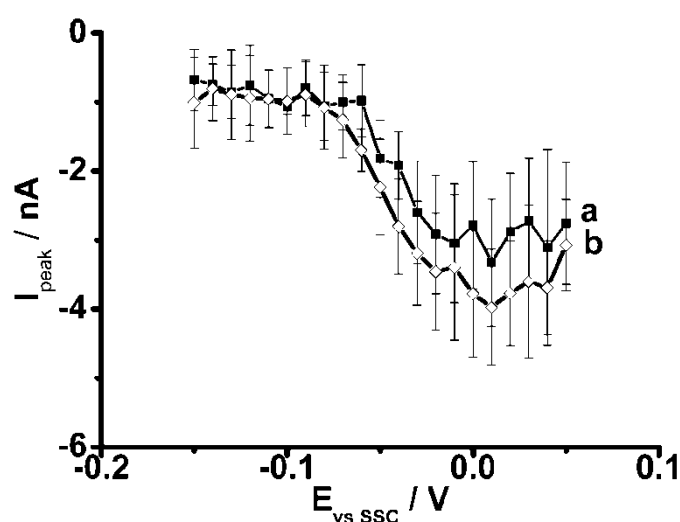


**Figure 4.16:** Effect of the concentration of the complexing agent (CAA) upon AdSV response in a) blank sample with  $2 \mu\text{g}\cdot\text{L}^{-1}$  of uranium (VI) and b) SoUV sample. AdSV condition is as in table 1.

#### 4.3.3. Pseudopolarography

In Fig. 4.17 we presented pseudopolarograms of U (VI) recorded for the 6hrs SoUV Sample (a) and the 6hrs UV Sample (b). The Original sample was not included in this comparison because the free U (VI) level in this sample was too low to be detected under the experimental conditions. The results of this experiment demonstrated that, both types of the UV sources (soft solar and hard artificial) produced dramatic effect upon the stripping response of U(VI) in the range between  $-0.15$  and  $0.05$  V deposition potential. Whereas the maximal achievable voltammetric signals for the UV Sample were not much larger as compared to the SoUV Sample, also the potentials shifted only slightly towards negative direction. The effects of solar UV irradiation treatment were much larger in case of U (VI) compared with our earlier studies of Zn, Cd, Pb, and Cu [Woldemichael 2011], as well as Ni

and Co [Sander 1999]. The rationale behind these phenomena can be explained in terms of complexation of the analyte cations by different matrix components that are differently prone to UV oxidation, and also form complexes of very different stability with the various analyte metal ions. Different interfacial activity of the dissolved organic compounds in river water samples can be the second reason. These may disturb the adsorptive accumulation of the analyte ion complexes as was observed with DMG for nickel and cobalt so does with CAA for uranium



**Figure 4.17:** Pseudopolarograms for uranium (VI) in a) 6 hours SoUV sample, b) 6 hours UV sample measured at the following experimental conditions: Initial deposition potential -0.15 V, final deposition potential -0.05 V scan increment 0.01 V at 0.01V.s<sup>-1</sup> scan rate. Scans were performed from 0.05 to 0.15 V. Other conditions were the same as in table 1.

**Table 4.7:** Recovery tests for uranium (VI) in Original, SoUV, UV Samples, and a blank sample.

Samples	Time of irradiation	Concentration ( $\mu\text{g}\cdot\text{L}^{-1}$ )			Recovered	Recovery rate (%)
		Spiked	Before Spiking	After Spiking		
<b>Original</b>	0hrs	1.00	0.00	0.00	0.00	0.00
<b>SoUV</b>	6hrs	1.00	0.88	1.81	0.93	93.00
	12hrs	1.00	1.03	1.97	0.94	94.00
<b>UV</b>	6hrs	1.00	1.07	2.01	0.94	94.00
	12hrs	1.00	1.12	2.07	0.95	95.00
<b>Blank</b>	0hrs	1.00	0.00	1.08	1.08	108.00

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In contrast to the earliest reports on the use of CAA in U (VI) AdSV [Sander 1994, Sander 1995], the samples of Warnow river water in our study absorbed and complexed nearly all U(VI) in the sample and the added  $100 \mu\text{g.L}^{-1}$ . Although the river water sample in the above reports was not described in detail, Warnow river water is found to carry high content of humic substances. We have reported about this earlier [Woldemichael 2011]. Other reports stated that UV sample pre-treatment may be necessary at higher DOC levels (above  $15\text{mg.L}^{-1}$ ) [Metrohm Application Bulletin No. 266/1, [www.metrohm.com](http://www.metrohm.com)]. Most reports in the literatures on AdSV determination of U (VI) in real samples focused on samples from tap, well or sea waters without major DOC contents. Metrohm states in its Application Bulletin that only tap water, ground water, sea and mineral water samples can be analyzed without UV digestion.

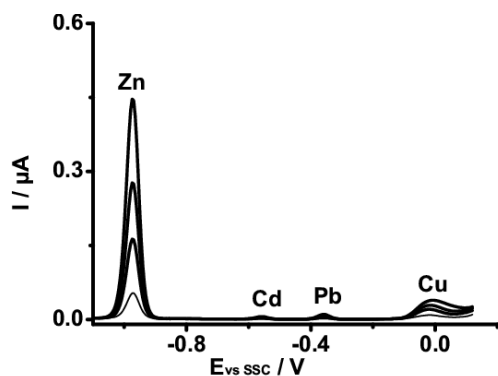
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#### ***4.4. Optimization of time and intensity of solar UV irradiation in natural water sample pretreatment steps for voltammetric trace metals determination.***

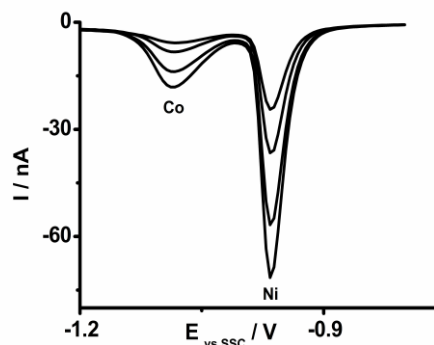
Awash River water samples were collected from Ethiopia Oromia regional state, East shewa zone at location given in table 3.2 on March 17, 2012. As a matter of fact, these locations are sub- equatorial regions with high Solar UV dose (averaged  $5.4\text{mW}/\text{cm}^2$  at the time of sampling). The altitude of this location ranges 1542-1595 meters above sea level. After the addition of  $\text{H}_2\text{O}_2$  (section 3.4), the acidified and filtered samples were Solar UV-irradiated in a thin UVA- transparent polyethene bag for 12 and 24 hours. All samples were packed and shipped to Rostock-Germany for voltammetric determination of trace metals repeating similar procedures as demonstrated by G. Woldemichael et al [Woldemichael 2011, Woldemichael 2012] for Warnow River water samples. Basically this experiment is aimed to achieve two objectives. These are:

a) Optimization of the time required to complete the digestion process of dissolved organic matter (DOM) in the river water samples at locations with sufficiently high UV dose like equatorial or tropical regions.

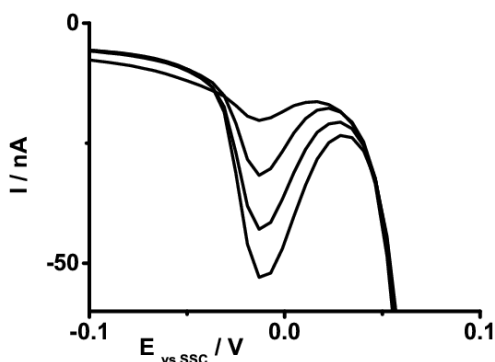
b) Determination of the concentration of the target heavy metals (cadmium, cobalt, copper, lead, nickel, uranium and zinc) in Awash River by applying the newly developed sample pre-treatment method in toxicological analysis of these trace metals. Accordingly, voltammetric determination of the 12hrs, 24hrs and the 24hrs SoUV + 6hrs artificial UV irradiated samples are carried out (figure 4.17a-c) based on the previously developed methods [Woldemichael 2011, Woldemichael 2012]. The numerical values of the concentrations calculated for 12hrs SoUV, 24hrs SoUV and 24hrs SoUV + 6hrs artificial UV irradiated samples are presented in table 4.8. The analysis of the values indicated the optimum time of solar UV irradiation required to bring about complete destruction of DOM in river water samples in such sub-equatorial regions and mid-altitude. The studies of the values confirmed that, 12hrs of solar UV irradiation at the above conditions could mineralize large portion of the matrices. However, the decomposition was incomplete and this was later confirmed by the results obtained after additional 12hrs of solar-UV irradiation and even extra 6hrs artificial UV irradiation with 30 W UV lamp of 254nm. This showed clearly additional 12hour Solar UV irradiation increased the concentration of almost all the metal ions dramatically.



**Figure 4.18a:** Typical Voltammograms for DpASV determination of Cd (II), Cu (II), Pb (II) and Zn (II) in Awash River.



**Figure 4.18b:** Typical Voltammograms for DpAdSV determination of Co (II) and Ni (II) in Awash River.



**Figure 4.18c:** Typical Voltammogram for DpAdSV determination of U (VI) in Awash River.

The amount of free metal ions available for voltammetric determination was increased by 16.99% for Zn(II), 66.67% for Cd(II), 37.91% for Pb(II) and 37.73% for Cu(II). Further treatment of the samples with artificial UV radiation for 6hrs showed no significant increment on the concentration of the metal ions under consideration (table 4.8). This reveals that 24hrs of solar-UV irradiation at average intensity of  $5.4\text{mW}\cdot\text{cm}^2$  or more will be sufficient for complete distraction of dissolved organic matter (DOM) in natural waters such as river water, sea water, lake water and so forth. In similar determination of the concentration of Ni(II), Co(II) and U(VI) by DpAdSV, the concentrations increased by 26.40% , 9.80% and 18.18 % after 24hrs of solar-UV irradiation. The confirmation of the total mineralization or the destruction of dissolved organic matter (DOM) was demonstrated by irradiating the 24hrs SoUV sample with artificial UV for 6hrs. The results of this experiment showed that, further UV irradiation has no significant effect on the amount of free metal ions available for

voltammetric determination. This would mean that, the sample was already undertaken complete digestion and no more dissolved organic matter was left for further treatment.

**Table 4.8:** *Effect of UV dose on the amount of available free metal ions as a consequence of the destruction of DOM in Awash River water samples collected from Sire Robi.*

UV Dose	Concentration ( $\mu\text{g.L}^{-1}$ )						
	Zn (II)	Cd (II)	Pb (II)	Cu (II)	Ni (II)	Co (II)	U (IV)
12 hours SoUV	115.89	0.12	3.06	26.61	4.66	0.51	0.18
24 hours SoUV	135.53	0.20	4.22	36.65	5.89	0.56	0.22
24 hours SoUV + 6 hours UV	137.85	0.20	4.24	37.01	5.97	0.55	0.22
% increment due to additional SoUV	16.99	66.67	37.91	37.73	26.40	9.80	18.18
% increment due to additional UV	1.02	--	0.47	0.98	1.36	--	--

This confirms that, 24hrs of solar-UV irradiation at the given intensity can be employed as sample pre-treatment step in the determination of trace Ni (II), Co (II) and U(VI) in river water or other natural water samples by the above instrumental technique. In summary, this experiment demonstrates that, solar-UV photooxidation at higher Solar-UV intensity (such as tropical or equatorial regions) can be used as a sample pretreatment method in DpASV and DpAdSV methods of trace metal determination. The method is economically affordable, environmentally friendly and clean. The materials we need in this experiment are sample holders, thin polyethylene bags small amount of  $\text{HNO}_3$  and  $\text{H}_2\text{O}_2$ . These materials and chemicals are locally available. Otherwise we can find them in most of the analytical laboratories.

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#### ***4.5. Toxicological review of Awash River in respect to cadmium, cobalt, copper, lead, nickel, uranium and zinc***

Prior to the determination of the concentration of the metals we determined the method detection limit (MDL) for each metal. This was the necessary step to evaluate the region where the signal to noise ratio (S/N) is greater than 5 as stated in the protocol and to check whether the observed values of the concentrations of each trace metal is different from noise because detection limit is matrix, method, and analyte specific. The values of the measured MDL are presented in table 4.9 in comparison with IDL recommended by the manufacturer of the instrument (Section 3.1). These values proved that all the detected metal ions concentrations are above the values that can be detected by this protocol and the methods we established in our previous works. Therefore, we applied the above method for practical application of the determination of the concentrations of seven heavy metals (table 4.9) in Awash River.

**Table 4.9:** Method detection limit (MDL) determined according to the procedure given in section 3.6.

<b>Analyte</b>	<b>IDL (ng.L<sup>-1</sup>)</b>	<b>MDL(ng.L<sup>-1</sup>)</b>	<b>Spike concentration(ng.L<sup>-1</sup>)</b>
Cadmium	50	73.00	100
Cobalt	50	89.00	100
Copper	50	260.00	100
Lead	50	240.00	500
Nickel	50	58.00	100
Uranium	25	120.00	100
Zinc	50	274.00	1000

The analytical results of the samples collected from different sites of Awash River are employed to evaluate the degree of contamination of the river in respect to the target heavy metals based on international (WHO) water quality standards. We also evaluated the status of health hazards (risks) associated to heavy metal pollution of Awash River. In general the determination of the concentration of these heavy metals helped us to analyze toxicological impacts related to this major heavy metals' contamination. The analytical results of the concentration of the heavy metals under consideration are summarized in table 4.10. The table includes background concentration derived from all possible contaminations such as reagent

grade HNO<sub>3</sub>, plastic bottle (sample holder), polyethylene bag, the filter paper, the cell, the buffer and so forth. Critical evaluation of the variation of the concentrations of the samples collected from Koka Dam and Sire Robi is demonstrated in table 4.10. This investigation reveals that, the concentration of Cu (II), Pb (II), Ni (II), U (VI) and Zn (II) increased downstream by 45.42%, 25.85%, 28.36%, 3.00% and 8.86% respectively. To the contrary, the concentrations of Cd (II) and Co (II) decreased downstream by 3.52 % and 4.84% respectively. As indicated in section 3.4 of this dissertation, Koka Dam samples are collected from the upstream of the Dam just 200 meters before the river joins Koka artificial lake. The time of sampling corresponds to the dry season at that locality implying low flow rate, low water volume and high DOM content. In the other side, Sire Robi is located downstream or beyond the outlet of the dam (about 3km from Koka hydroelectric power station) just at Adama drinking water treatment plant. The rationale behind the decrease / or increase in the concentration of metal ions downstream can be explained based on various factors. For instance, the physical and chemical processes taking place in the water, the velocity(flow rate) of the water, turbulence, water volume, landscape, aquatic fauna and flora are some of the factors that affect migration of the metal contaminants and the matrices or the fate and transport of the complexing DOM and hence the metal ions. In this particular case, the velocity and turbulence of the water is decreases quickly as soon as the river inters the lake.

**Table 4.10:** comparison of percentage variation of the concentration of heavy metals of two sampling sites

Sampling Site	Concentration (µg.L <sup>-1</sup> )						
	Zn	Cd	Pb	Cu	Ni	Co	U
Koka Dam	126.62	0.21	3.37	25.45	4.67	0.57	0.19
Sire Robi	137.85	0.20	4.24	37.01	5.97	0.55	0.22
Concentration Variation (downstream) %	8.86	-4.84	25.85	45.42	28.36	-3.52	3.00

This will give additional chance for DOM to interact with other anions and cations as a result of ion exchange process which might help sedimentation of the matrices together with the complexed metal ions. There are also other sinks like plants and animals inhabited in the lake which may consume considerable quantity of the metal ions. In other words the lake serves as a natural sink of trace metals. As a result the concentration of Cd (II) and Co (II) decreased at the outlet of the water from the dam. This corresponds to the expected outcome



downstream. To the contrary, the concentrations of Cu (II), Pb (II), Ni (II), U (VI) and Zn (II) increased downstream in astonishing extent. This unexpected increase in concentration might have several root causes. Some of the possible causes for the pollutions are suggested as follows.

**Table 4.11:** Variation of the concentration of heavy metals downstream of Awash River

Element	Background conc. ( $\mu\text{g.L}^{-1}$ )	Background corrected concentration ( $\mu\text{g.L}^{-1}$ )				
		Awash Malkasa	Wonji Drain	Wonji Kuruftu	Sire Robi	Koka Dam
Zn	28.09+0.86	440.85+11	26.12+2.62	76.70+7.11	137.85+1.60	126.62+1.13
Cd	0.00	2.87+0.05	0.06+0.02	0.11+0.03	0.20+0.01	0.21+0.03
Pb	1.59+0.05	10.93+0.50	0.52+0.05	1.14+0.21	4.24+0.09	3.37+0.17
Cu	6.25+0.72	4.33+0.06	11.70+0.80	25.71+2.27	37.01+3.72	25.45+0.56
Ni	2.21+0.01	0.75+0.09	3.35+0.51	20.44+0.29	5.97+0.94	4.67+0.45
Co	0.00	0.00	0.21+0.06	0.00	0.55+0.03	0.57+0.04
U	0.84+0.00	1.41+0.17	0.56+0.47	1.35+0.21	0.22+0.13	0.19+0.25

One of the most possible causes can be the interaction of the water with metal components, lubricants and other chemicals such as paints at the hydroelectric power station. The surrounding soil and agrochemicals used by the local farmers could be the other source of contamination. The nearby Tannery (Ethiopian Tannery) also discharges its waste to the lake. This deadliest waste can contribute to the unexpected sharp increase in the concentration of these metals. In addition to these, there are extensive horticultural fields which apply agrochemicals intensively. The leaching of these chemicals to the lake could lead to the increase of the concentration of the metals there by polluting the river water and of course the environment and the ecosystem. However the reason why the concentration of the metal pollutants increased downstream requires further investigation to identify the point and non-point sources and to recommend possible solutions. Further investigation also revealed that some metal ions increased in concentration much more than expected while others decreased downstream. For instance Zn (II) increased from  $126.62\mu\text{g.L}^{-1}$  to  $440.85\mu\text{g.L}^{-1}$  as we move from Koka dam to Awash Malkasa. To the contrary, Cu (II) concentration decreased from  $25.45\mu\text{g.L}^{-1}$  to  $6.25\mu\text{g.L}^{-1}$  for the same locations. In general the determination of the concentration of these heavy metals helped us to analyze the degree of pollution and hence their toxicological impact on humans and animals based on the WHO safe drinking water guideline quality standards (table 4.12). Deep investigation of the concentration data of the metal ions in the river would lead to the conclusion that, it is unsafe for drinking in respect to

lead, while it is in the range of safe drinking water in respect to cadmium, copper, nickel, uranium and zinc. In conclusion, local people living around Awash River and the Adama City dwellers who drink Awash River are at risk of lead poisoning and the health hazards that accompany it. We have also observed that, the concentration of cobalt is alarmingly increasing downstream and it needs serious attention. We suggest further studies to identify the root causes of the contamination and to recommend remedies.

**Table 4.12:** Heavy metals' pollution status in Awash River according to World health organization (WHO) Guidelines for Drinking-Water Quality, Geneva,

<b>Element</b>	<b>Lever in Drinking water (<math>\mu\text{g.L}^{-1}</math>)</b>	<b>Maximum observed concentration (<math>\mu\text{g.L}^{-1}</math>)</b>	<b>WHO Guideline Value (<math>\mu\text{g.L}^{-1}</math>)</b>	<b>Toxicological evaluation</b>
Cadmium	1.0	2.87	3.0	Endangered
Cobalt	Not specified	0.57	Not specified	Unknown
copper	5 – 30,000	30.76	2000.0	safe
Nickel	<20.0	10.93	70.0	safe
Lead	5.0	20.44	10.0	unsafe
Uranium	<1.0	1.41	15.0	Safe
zinc	10.0 – 50.0	440.85	3000.0	Safe

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## 5. Summary and Outlook

This study demonstrates that solar irradiation assisted by low pH and addition of hydrogen peroxide can serve as a means for water sample digestion. The irradiation time can exceed 24 hours at mid latitudes while 24 hours are enough at equatorial or tropical regions as demonstrated in this study. The Solar UV intensity can be increased by using a kind of solar collector such as aluminum reflector. Cheap UV transparent materials such as PET bottles or thin polyethylene bag serves as sample holder during irradiation. The presented protocol can be recommended for ASV determinations of trace elements such as cadmium, copper lead and zinc in river water any natural water samples at remote places in the environment or otherwise, where electric high-power UV lamps or commercial UV digestion units cannot be used. The application can be extended to AdSV determination of trace metals by employing hydrogen peroxide-assisted irradiation of river water samples with solar UV radiation. This was proved by the determination of nickel and cobalt on hanging mercury drop electrodes. The effect of UV irradiation treatment was even larger compared to anodic stripping voltammetry of the above heavy metals (Zn, Cd, Pb, and Cu) in the same river, as reported earlier. We also investigated that Solar-UV digestion is applicable for AdSV ultratrace determination of uranium (VI) in river water. An exposition of 12 hours under Central European conditions yielded the same digestion effect as compared to hard artificial UV radiation from a 30 W, 254 nm sources. We also found that certain river waters with exceptional high organic content can completely block the AdSV signals of U (VI). These organic components can complex or absorb up to  $100 \mu\text{g.L}^{-1}$  of added uranium hindering or distorting voltammetric signals. The effect of solar UV irradiation treatment was even larger compared to anodic stripping voltammetry of Zn (II), Cd (II), Pb (II), Cu (II), as well as AdSV of Ni (II), Co (II) and U (VI) in samples from the same river, as reported earlier. The approach reported here should be very useful for mobile electrochemical heavy metal analysis in regions where frequent sunny weather conditions are available as proved in the determination of Awash River water samples at low latitude-Ethiopia. The method is applicable in practical and real sample analysis for its heavy metals pollution or contamination. It is also useful in the analysis of toxicological and ecotoxicological aspects of heavy metals in natural waters such as river water, sea water, lake water and so forth by ASV/AdSV methods as demonstrated in this work (the case of Awash River-Ethiopia). In this

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study we concluded that, Awash River is unsafe for drinking in respect to lead, while it is in the range of safe drinking water in respect to cadmium, copper, nickel, uranium and zinc. In addition to these the concentration of cobalt is increasing downstream in alarming size and it needs serious attention by the stake holders.

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