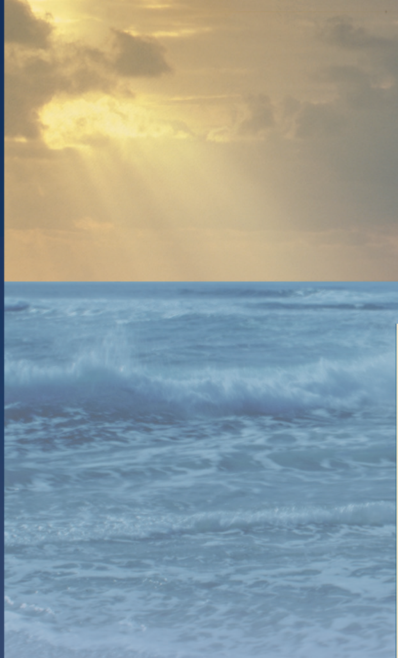

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Preface

This report is a contribution to the ERMS task addressing toxicity of drilling discharges. The report addresses a specific sub-task within the toxicity task, carried out by RF-AM. The report is a contribution to the work carried out by the other project participants. The main activity is led by TNO.

The report addresses primarily leaching, uptake, bioavailability and effects from metals, and is partly based on the experimental data genereateed in RF-AM through other project activites working with historic drill cuttings discharges.

Work participants:

Thanks to contributors:

Key Words:

Metals, leaching, uptake, bioavailability, effects, partition factors

Stavanger (19/11-2004)

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Introduction

This report is a contribution to the ERMS task addressing toxicity of drilling discharges. The report addresses a specific sub-task within the toxicity task, carried out by RF-AM. The report is a contribution to the work carried out by the other project participants. The main activity is led by TNO.

ERMS: Metals from drilling discharges

Three aspects are of particular relevance in assessing the impacts caused by metal contamination in drilling discharges:

- metals leaching
- metals bioavailability
- effects from metals to biota

Each of these issues are addressed in the following.

Introduction –drilling discharges

Drilling discharges spread over large areas (Rye et al., 1998, Muschenheim and Milligan, 1996), and stay in the water column for a prolonged time, and the potential for impacts are thus considerable given the volumes and suit of components being discharged. Through the UKOOA studies (Westerlund et al. 2001, Kjeilen et al., 2001, see also UKOOA web-site: www.oilandgas.org.uk) it was established that cuttings piles may be affected by storm incidents down to depths of 100 m (Sabeur et al. 2002). Studies have also shown that erosion of cuttings piles may be a significant process (Vefsnmo and Lothe, 2001), resulting in re-suspension and spreading in the water column. Hence, both pelagic and benthic organisms can be repeatedly exposed, both by "primary" exposure as the material settle through the water column and as "secondary" exposure due to resuspension and repeated settling of particulate material.

To meet required mud design criteria, drilling weight materials (such as barite and ilmenite), comprising up to 90% of the mud, are used as small particles. The barite and ilmenite used are grained into small particles of specific grain sizes for use as components in drilling formulations, particle sizes being in the range 0.0007-0.05 mm, with a typical diameter of 15-20 micrometer. These weighting agents are used in large quantities, usually in a volume 1-5 times larger than the volume of cuttings from the well (Kjeilen et al. 2001). Barite is to various extents contaminated with metals, one of the major contaminants being lead. Ilmenite generally contains lower concentrations than barite of metals of environmental concern. Other sources of metal contamination can also be present in the mud (drilling formulation).

To elucidate the true impacts from drilling discharges, it is important to consider what biological resources are the most relevant. It is suggested to focus on most sensitive organisms/functional groups, which will/might be either:

- regional
- seasonal
- growth stage dependent

As an example, e.g. cod larvae (Gadus morhua) are often more sensitive to contaminants than juveniles or adults (Hutchinson et al., 1998).

Leaching

In the UKOOA drill cuttings project (Westerlund et al. 2001) a sequential extraction procedure (modified after Tessier and Champell, 1979 by Lopez-Sanches et al, 1992) was applied to cuttings collected from 12 different cuttings deposits in the North Sea in order to investigate leaching properties of the deposits. These leaching studies of metals from historic cuttings material showed that the elements Zn, Cu, Cd and Hg were found in fractions loosely bound to particles, thus indicating that these elements have a potential to accumulate in biota.

In the Norwegian Research Council (NRC) funded pre-project “impacts of metals from drill cuttings and mud to the marine water column” (2002) the same methodological approach was applied to different drilling muds and components, intending to identify important processes, such as impacts from ageing, weathering, biological transformation etc., responsible for the mobilisation of metals from discharged cuttings and mud in connection with the drilling activities (Westerlund et al., 2002). In this study it was shown that certain elements were more easily extractable in barite-based muds compared to ilmenite-based muds. Likewise, metals seemed to be more easily extracted from water based mud (WBM) as opposed to oil-based muds (OBM). Metal levels in total also were generally higher in barite-based formulations. This is exemplified in the figure below. Again Zn, Cu, Cd and Pb have the highest contribution in the leachable fraction (Westerlund et al., 2002). Also, it was shown that Cu and Zn were more available from barite and barite-based formulations than from a “natural” sediment, while other elements were less available compared to an average sediment. Likewise, Ni were more available from ilmenite compared to natural sediment while other elements were less available. The concentration of Ni in ilmenite is rather high.

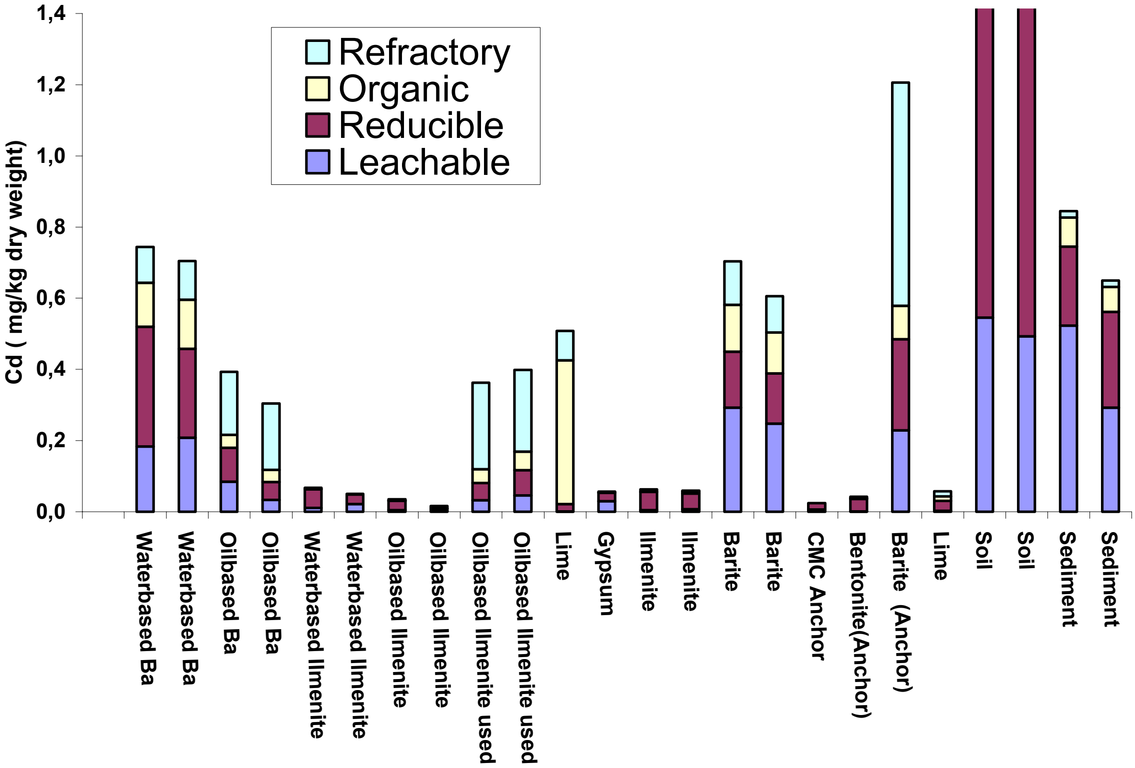

Figure 1. Levels of Cd in various designed muds and single drilling formulation components (from Westerlund et al., 2002).

Metal partition coefficients calculated from sequential extraction studies

Partition coefficients calculated based on the two above mentioned studies and some literature data is presented below. It must be emphasised that these partition coefficients are derived from data sets evolved from chemical extraction only, thus not taking into account physical and biological processes which will dictate a more realistic leaching scenario.

The partition factors in Table 1 are calculated as the part of the selected metal in the most mobile extraction fraction. The sediment samples represent both literature reported data (Filgueiras et al., 2002, Lopez-Sanches et al., 1993) applying the same sequential extraction procedure applied for the cuttings and drilling chemicals (Westerlund et al., 2002, Westerlund et al., 2001). The cuttings material from a number of cuttings piles are included, and divided into two groups.

Table 1. Partition factors calculated from the studies by Westerlund et al. (2001, 2002) and more. 'Cuttings High' represent cuttings samples with a high metal content). 'Cuttings Low' represent cuttings pile material with a lower metal content).

Partition factors

	Cr	Ni	Cu	Zn	As	Cd	Pb
Sediment	0.40	0.40	0.20	0.50	0.50	0.85	0.40
Barite	0.30	0.30	0.50	0.70	0.10	0.70	0.20
Ilmenite	0.30	0.80	0.10	0.30	0.80	0.90	0.30
Cuttings High	0.20	0.40	0.05	0.20	0.05	0.20	0.20
Cuttings Low	0.50	0.40	0.10	0.30	0.15	0.30	0.30

Partition factors normalised to sediment

	Cr	Ni	Cu	Zn	As	Cd	Pb
Barite	0.75	0.75	2.50	1.40	0.20	0.82	0.50
Ilmenite	0.75	2.00	0.50	0.60		1.06	0.75
Cuttings High	0.50	1.00	0.25	0.40	0.10	0.24	0.50
Cuttings Low	1.25	1.00	0.50	0.60	0.30	0.35	0.75

These factors can be used for risk assessment given that the total concentration of metals (NS 4770 Digestion procedure) in the mud/drilling discharge is known. With these weighing factors the discharged mud can be evaluated as a sediment. When assessing the risk in the water column the barite and ilmenite data could be used (then assuming more spread of the particles). When assessing the risk in the sediment the cuttings data should be used (e.g. once settled on the seabed the material are more similar to that represented by the cuttings high and low data above).

In summary, the following observations were made:

- New WBM mud with either barite or ilmenite gave similar results as pure barite and ilmenite, the mixing of barite and ilmentite into a mud thus did not affect the availability of the metal elements. Hence, partition factors derived for the pure compounds can be applied in risk assessments
- Metals from cuttings accumulations on the seabed (old, used drilling mud and rock cuttings) were in general less available than in a "normal" sediment. Also, the relative availability of the metals decreased in more contaminated cuttings material.

Bioavailability

Bioavailability describes the portion of a contaminant that can be taken up by the organism from its environment and food and is subsequently transported, distributed and metabolized by the organism (Kördel et al., 1997). Both uptake and bioavailability of metals are important measures in assessing impacts. Little is known about the potential long-term effects of suspended particles from drilling mud discharges on water-column organisms. There is a general belief that water based drilling discharges pose little environmental risk to organisms in the water column. Such believes are partly founded on the ongoing process of testing acute toxicity of single chemicals (OSPARCOM tests), but toxicity testing of drilling formulations does not always match up with field observations, probably because (assumed) chemically inert material such as barite are not routinely included in such tests (Barlow and Kingston, 2001). It is generally assumed that the dissolved fraction of a toxic substance in surface water is mainly responsible for toxicity to aquatic organisms (Weltens et al., 2000). Weltens *et al*, however, showed that the particle bound fraction of metals can become available within the body of filter feeding *Daphnia* (Weltens et al., 2000). Adsorbed metals might desorb in the gastrointestinal tract due to different physico-chemical conditions and exert toxic effects and lead to unexpected high tissue concentrations (Weltens et al., 2000). The digestive physiology of the animal and the behaviour of the chemical within the animal's gut influence contaminant assimilation (Wang and Fisher, 1999a). For suspension feeders such as mussels and copepods, uptake of metals from the dissolved phase and food ingestion can be equally important to metal accumulation (Wang and Fisher, 1999b). Particle bound contaminants can also be bioavailable to fish (Qiao and Farrell, 1996; Van den Belt et al., 2000).

The bioavailability of Cr and Fe bound to colloidal microparticles or macromolecules to the green mussel *Perna virdis* and the clam *Ruditapes philippinarum* was examined by Pan and Wang (2002). Colloidal particles are defined as the size fraction between 1kDa and 0.2micrometer and are thus generally included in the dissolved phase of seawater (defined as <0.2 or 0.45 micrometer). Colloidal particles are often of organic origin and are generally much smaller than average drilling discharge particles. The findings of Pan and Wang (2002) are however interesting in that they addresses bioavailability in a manner that may be of relevance also with larger particles.

Pan and Wang (2002) found that uptake of the colloidal bound metals was much higher than uptake from low molecular weight complexed metals, and colloid-bound metals were accumulated by the bivalves. The difference was largest with Cr. Metals bound with a large size colloid fraction (10kDa -0.2 micrometer) were more bioavailable than metals bound with smaller size colloids (<10kDa). Pan and Wang (2002) refer to a number of studies addressing bioavailability of colloid associated metals to mussels, clams (Wang and Guo, 2000; Cd, Cr, Zn), shrimp (Carvhalo et al, 1999), American oyster, *Crassostrea virginica* (Guo et al, 2001), zebra mussels (Roditi et al, 2000; Cd, Ag, Hg) and marine phytoplankton (Nodwell and Price, 2001). Metals associated with organic colloids have been shown to be more bioavailable than truly dissolved metals or metals bound to lower molecular weight fractions (Pan and Wang, 2002, Roditi et al. 2000, Guo et al. 2001). Whether the same is true for inorganically bound colloidal particles has not been addressed. A considerable fraction of metals taken up as colloids was distributed in the bivalve soft tissue after exposure (Pan and Wang 2002). Up to 60% (Cr) and 70% (Fe) were found in the digestive glands after 1-4h exposure, further implying the possibility of direct ingestion of colloidal particles by bivalves, which may be partially responsible for the high bioavailability of colloid-bound Cr and Fe to the bivalves. It is stated (Pan and Wang, 2002) that the chemical "quality" and origin of colloids can have substantial influence on the bioavailability of colloid-bound metals to both mussels and clams, but the mechanisms underlying these differences remain to be revealed.

In testing whether mussels (Mytilus edulis) could be recommended for use as an indicator for metal contamination (Zn, Cd, Cu and Pb), the influence of water temperature, salinity, seasonal variation, position of specimen in the water column etc. was found to have little influence on metal uptake (Phillips 1976). Also, the simultaneous exposure to all metals did not adversely affect uptake in the mussels. The exception was Cu that was influenced by all parameters tested (salinity, temperature, simultaneous exposure).

Oomen et al (2003 a and b) investigated the uptake of lead in humans, especially intake of contaminated soil in children, which might be a major exposure route. The uptake was examined in relation to speciation of the lead, results indicating that not only free Pb^{2+} but also other forms of lead (e.g. lead phosphate) are available for transport across the intestinal epithelium. This work then suggests that various forms of metals may be bioavailable. No references have been identified addressing whether different species of lead are accessible in marine organisms.

There are a number of publications addressing methods of sequential leaching/extraction etc. to measure bioavailable fraction and metal species etc. These references contain no information on actual sizes of bioavailable fractions from given environmental samples of use in assessing metals leaching and bioavailability from cuttings material.

Effects

The effects of drilling discharges have been investigated primarily for the benthic fauna, such as through the regular Norwegian monitoring programme. Offshore monitoring shows a link between environmental concentrations of total hydrocarbons (THC) and fauna composition/status. But, there is also a link towards Ba and Sr and to a lesser extent towards Zn, Cu, Cd and Pb (Gray et al., 1999, Olsgard and Gray, 1995).

Results from the NFR pre-project (Westerlund et al., 2002) studies show that mud from barite proved significantly more toxic than mud using ilmenite as the weight agent. Since ilmenite contained significantly lower levels of leachable heavy metals than barite, the results may indicate toxic effects of dissolved metals.

Suspended clay either from natural sources or from drilling mud can interfere with suspension feeding and damage the gills of bivalves (Morse, 1982, Stevens 1987, Cranford & Gordon, 1991, 1992) and fish (Sprague and Logan, 1979). Chronic intermittent exposure of sea scallops *(Placopecten magellanicus)* to dilute concentrations of operational drilling wastes, characterised by acute lethal tests as practically non-toxic, can affect growth, reproductive success and survival (Cranford et al., 1999). Negative effects were observed at 0.5 mg/L barite. The gills of the suspension feeder *Cerastoderma edule* and the deposit feeder *Macoma balthica* were damaged when exposed to suspended barite particles (Barlow and Kingston, 2001). Levels of barite accumulation that can be expected 100-500 m from a point of active drill cuttings discharge, caused 100% mortality within 12 days.

The copepod Acartia tonsa is a suspension feeder, and hence may be affected by exposure to suspended particles from the drilling mud. Copepods *(Acartia spp)* feeding on algae exposed to metals (Cd or Hg) experienced decreased egg production, hatching rate, ovarian development and egg protein content, implying that the process of yolk accumulation (vitellogenesis) was affected. Less effects were observed following exposure to the same metals in dissolved form (Hook and Fisher, 2001). Although the bioavailability of metals from the drilling mud particles may be considerably lower than from algae, these results show that particle bound metals can cause effect in copepods.

The larvae of mussels *(Mytilus edulis)* are filter feeding planktonic organisms. Toxic compounds (including metals) can affect survival, development and growth of mussel larvae (e.g. Beiras and His, 1995; Hansen et al., 1997; His et al., 2000; Hoare et al., 1995; Wedderburn et al., 2000b).

Intuitively it is difficult to separate between effects caused by particles or by metals released from the particles inside the organism. So far it has not been established whether observed effects are caused by the particles themselves or by the metal contaminants on them. There are several examples in the literature of studies showing various types of effect/responses to different effect parameters, as presented in the following.

Since metals accumulated from particles (e.g. barite) will induce metal binding proteins, one approach would be to examine metallothioneins (MT) to decide whether effects are caused by metals released from the particles inside the organism (on gills/in the gut), or because of physical effects of particles. Metallothioneins are small Cys-rich proteins which bind transition metals, and they play a significant role in the detoxification of pollutant metals and are clearly induced in metal-exposed marine invertebrates (Lopez-Barea and Pueyo, 1998) and e.g. Beyer et al., (1997); Wu and

Hwang, (2003). Metallothionein-like proteins have been detected in mussels e.g. (Amiard-Triquet et al., 1998; Blackmore and Wang, 2002; Bolognesi et al., 1999), copepods (Barka et al., 2001), and mussel larvae (Geffard et al., 2002).

Physical stress from exposure to suspended barite, and/or effects of metals that may leak from barite particles, and chemicals adsorbed to the particles may damage the gills of cod and bivalves. Waterborne metals can bind to gills of fish and disrupt the ionoregulatory and respiratory functions of the gills (Playle, 1998). Histopathological changes have been observed in gills from metal exposed fish (Muhvich et al., 1995; Thophon et al., 2003) and bivalves (Clark et al., 2000; Teh et al., 1999).

Further, effects of particles on the respiratory function and/or toxic effect of metals/chemicals in a drilling mud may cause oxidative stress in bivalves and cod. Oxidative damage has been causally linked to various kinds of environmental stress, both natural and artificial, the result being impairment of fundamental cellular functions like DNA (Frenzilli et al., 2001). Exposure to metals has been related to increased production of reactive oxygen species (ROS) (Muhvich et al., 1995;, Almeida et al., 2002).

Evidence of long- term adverse effects, such as cancer, due to heavy metals in marine animals has been shown in field and experimental studies (Bolognesi et al., 1999). Genotoxic effect may be involved in the mechanism of metal carcinogenicity. Methods such as the comet assay (developed by Singh et al., 1988) has been used to detect DNA damage caused by metal exposure of fish (Risso-de Faverney et al., 2001) and mussels (Bolognesi et al., 1999); (Black et al., 1996). The comet assay has been established in the RF-Akvamiljø laboratory for mussels, sea urchins and fish (Taban et al., 2003; Bechmann et al., 2002).

To assess the effects of suspended particles in the drilling mud on the bioavailability of metals, suspended matter can be considered as metal-binding ligands (Karman *et al*., 1999). The strength of the binding capacity, and its implications on uptake and effects in biota, can be predicted by adjusting modelling tools that are developed to predict metal speciation related to water quality parameters (Paquin et al., 1999). In assessing the environmental risk of metals, including essential ones, care has to be taken on the risk assessment methodology (Karman and Jak, 1998).

Strømgren, (1982) studied short-term (10-22days) effects on length growth of mussels (*mytilus edulis*) exposed to soluble metals (Zn, Hg, Cu, Cd, Pb and Ni) in seawater. The intention was to establish threshold levels of toxicity. EC50 values observed were 0.3-0.4 microgram/l Hg, 3-4 microgram/l Cu, 60 microgram/l Zn and 100 microgram/l Cd. Pb and Ni had no effect on growth up to a concentration of 200 microgram/l. Metal concentrations reported are the nominal concentrations added to the seawater. However, due to organic and inorganic complexation the concentration of toxic metal species available to the organism may be considerable less. The EC50 value of Hg is only one order of magnitude higher than concentrations found in seawater from the northern hemisphere according to Florence and Batley (1980). The levels for Hg are however more than 100 times higher than normally observed in the North Sea (Westerlund, pers. comm.).

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