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# EXPERIENCE OF BIODEGRADATION FOR THE DISPOSAL OF WASTE MACHINE TOOL CUTTING FLUID

G T Taylor and V C M Freestone
AWE [a], Aldermaston, Reading, RG7 4PR, UK.
Tel: 0118 9827551 Fax: 0118 9815320

## ABSTRACT

The disposal of waste cutting fluid from the machining of uranium is problematical, because the presence of oil in the cutting fluid causes difficulties for either aqueous or solid radwaste treatment. Biodegradation offers the potential to convert this material into forms amenable to disposal as low-level radioactive waste. The real bonus of biodegradation crucially depends on the degree of mineralisation achieved, because any by-product of biodegradation other than carbon dioxide will require disposal as secondary radioactive waste.

The initial work to investigate the benefits of biodegrading the particular cutting fluid used at AWE was reported at WM99 (1). At that time only about a third of the organic material was mineralised to carbon dioxide: the remainder was converted to microbial biomass and other residual organic material.

The present paper describes the further work undertaken to formulate a waste disposal process based on biodegradation. One involving ultrafiltration of the biotreated fluid followed by adsorption onto activated charcoal was examined. This process would produce a waste stream that would qualify as aqueous radioactive waste. It was shown that separated biomass could be immobilised in a cement matrix that would qualify as solid radioactive waste. Different conditions for biodegradation have been examined, but it was not possible to increase the degree of mineralisation of organic material to carbon dioxide. It was estimated that with the degree of mineralisation achieved, biodegradation would produce more solid waste than direct solidification of the untreated, waste cutting fluid in cement. Thus, at present biodegradation is not competitive. The authors propose additional work to optimise the biodegradation process and to examine alternative stabilisation methods in order to reduce the amount of solid secondary waste.

## INTRODUCTION

AWE's role to maintain the UK's nuclear deterrent involves machining uranium components, which generates waste cutting fluids contaminated with the radioactive metal. The machine tool cutting fluid that has been used at AWE for many years contains mineral oil, anionic and non-ionic surfactants, and a phenolic biocide ("cresylic acid"). At present the disposal of radioactive waste containing oils is

problematical, because the presence of oil causes difficulties for either aqueous or solid radwaste treatment. A legacy of about $10m^3$ spent cutting fluid has accumulated (2). Possible treatment processes, including biodegradation, chemical destruction and absorption for immobilisation in cement, are being investigated, with the aim of converting waste oil to a form that can be either treated as aqueous waste or disposed of as solid waste in the UK's repository for low-level waste (LLW) at Drigg in Cumbria.

Biodegradation of spent cutting fluid has been investigated (1, 2) because it has the potential to convert the oil and other organics to carbon dioxide, and so enable the cutting fluid to be treated along with the other aqueous radioactive waste generated at AWE.

This paper describes the experience gained at AWE of this application of biodegradation. The constraints on the application of biodegradation to the disposal of organic radioactive waste are different to the constraints on other industrial or environmental applications of biodegradation. The differences arise because any products from a biodegradation process with radioactive materials will have to be treated as radioactive waste. The disposal of carbon dioxide would be straight forward, because it could be filtered and discharged to the atmosphere in a monitored and regulated manner. However, the disposal of biomass or any intermediate break-down products would pose the same problems as the disposal of the original cutting fluid. Therefore, the potential for biodegradation of radioactive waste was seen to depend crucially on the extent of mineralisation to carbon dioxide. Consequently, the studies at AWE paid particular attention to the proportion of carbon dioxide in the carbon-mass balance (1). The author was unable to locate in the scientific literature any reports of a similar approach being taken to the evaluation of the performance of oil-wastewater treatment plants. It was reasoned that most studies of oil biodegradation have concentrated on the production of a liquid effluent suitable for discharge to a water course or a sewage works, where the main criterion for success is that the treated effluent meets any aqueous discharge limits.

## MATERIALS AND METHODS

### Bioreactor

The bioreactor installed at AWE was an Applikon Bioclave 110, equipped for monitoring dissolved oxygen (Broadley James' Oxyprobe model 20 dissolved oxygen transmitter), pH and temperature (Broadley James' model 10 pH transmitter). The contents of the bioreactor were stirred at 200 rpm and aerated through a sparge bar with 5 litres air/minute. The concentration of carbon dioxide in the outlet gas was measured using either a Deltagas infra-red carbon dioxide monitor (range 0 - 0.5%) or an Edinburgh Sensor's Guardian Plus infra-red carbon dioxide meter (range 0 - 5%),. Biodegradation was performed at ambient temperature, which was generally around 20 - 25 °C, and neutral pH (between about 7 and 8).

Several runs were performed in the bioreactor under different conditions (Table I). The results from the first 6 runs were displayed at WM'99.

Table I. Conditions for the Various Trials in the Bioreactor

| Run | Conditions |
|---|---|
| 1 | Indigenous Microbes , ~ 0.5 % Cutting Fluid, Bioreactor Stirred, Demineralised Water |
| 2 | Indigenous Microbes , ~ 2 % Cutting Fluid, Bioreactor Stirred, Demineralised Water |
| 3 | Indigenous Microbes , Cutting Fluid from various old stocks, Bioreactor Stirred, Demineralised water |
| 4 | Indigenous Microbes , ~ 0.1 % Cutting Fluid, Bioreactor Stirred, Tap water |
| 5 | Indigenous Microbes , ~ 0.1 % Cutting Fluid Added to Run 4, Bioreactor Stirred, Tap water |
| 6 | Indigenous Microbes , ~ 0.3 % Cutting Fluid, Bioreactor Stirred, Tap water |
| 7 | Indigenous Microbes , ~ 0.1 % Cutting Fluid, Bioreactor Not Stirred, Tap water |
| 8 | Indigenous Microbes , ~ 0.1 % Cutting Fluid, Bioreactor Not Stirred, Tap water |
| 9 | Non-Indigenous Microbes From Commercial Sources, ~ 0.3 % Cutting Fluid, Bioreactor Stirred, Tap water |
| 10 | Non-Indigenous Microbes From Commercial Sources,, ~ 2 % Cutting Fluid, Bioreactor Stirred, Tap water |
| 11 | Back to Indigenous Microbes, ~ 0.3 % Cutting Fluid, Bioreactor Stirred, Tap water |
| 12 | Indigenous Microbes, $H_2O_2$ Pre-treatment of ~ 0.3% Cutting Fluid, Bioreactor Stirred, Tap water |

**Growth of Indigenous Micro-organisms**

The inoculum of indigenous micro-organisms for the trials shown in Table 1 was derived from several stocks of used, non-radioactive cutting fluid held on-site. The inoculum for Figure 1 was grown in waste cutting fluid diluted to the equivalent of approximately 0.3 % w/v neat product in tap water and supplemented with sodium nitrate ($NaNO_3$) (3.64 g / litre) and potassium dihydrogen phosphate ($KH_2PO_4$) (0.29 g / litre). The growth medium was dispensed in conical flasks (to 10% of the volume of the flasks), which were incubated on a Gallenkamp/Sanyo orbital shaker at 100 rpm and at ambient temperature. This inoculum (1.7 litres) was added to the bioreactor, which contained 3.9 litres of waste

cutting fluid, 291 g $NaNO_3$ and 23.2 g $KH_2PO_4$, with the volume being made up to 80 litres with tap water. The concentration of cutting fluid in the bioreactor was equivalent to approximately 0.3% of the neat product. The inocula of indigenous micro-organisms used for other trials were grown in a similar manner or they were derived from the previous run in the bioreactor.

## Microbes from Different Sources

The properties of the "non-indigenous" microbes examined as part of this study are shown in Table II.

Table II. Sources of Microbes Used for Runs 9 and 10

| Culture / Source | Culture Collection Code | Biodegradative Properties * |
|---|---|---|
| *Acinetobacter* sp. | NCIMB 11507 | Degrades iso-alkanes |
| *Acinetobacter* sp. | NCIMB 11742 | Utilises crude oil |
| *Nocardia coeliaca* | NCIMB 9574 | Degrades asphalt |
| *Pseudomonas putida* | NCIMB 9816 | Utilises anthracene |
| *Pseudomonas putida* | NCIMB 10015 | Degrades cresol |
| *Pseudomonas* sp. | NCIMB 10750 | Degrades plasticised PVC |
| *Rhodococcus* sp. | NCIMB 11997 | Degrades alkylbenzenes |
| EcoSafe HD<br>- *Arthrobacter sp.*,<br>- *Bacillus sp.*,<br>- *Nocardia paraffinae* | | A proprietary blend of microbes marketed by BioIndustries, Dublin, as a microbial supplement for aerobic, biological wastewater plants treating oily wastes. |
| Undefined culture from the waste water treatment plant operated by Lubrizol, Cheshire | | Culture from aerobic, biological wastewater treatment plant used to treat water containing oils |

** Properties of the bacteria are taken from the catalogue of the National Collection of Industrial and Marine Bacteria.*

The pure cultures of bacteria were obtained as freeze-dried cultures from the National Collection of Industrial and Marine Bacteria (NCIMB) in Aberdeen. They were resuspended in 2 x 10 ml Nutrient Broth (Oxoid, prepared according to the manufacturer's instructions) and they were resuscitated by incubating the broths at ambient temperature for 3 - 4 days. In order to encourage the bacteria to express biochemical activities that might be needed to degrade the cutting fluid, they were sub-cultured into a mixture of Nutrient Broth and cutting fluid. Each broth culture (1 ml) was transferred into separate 500 ml conical flasks containing 10 ml nutrient broth and 40 ml cutting fluid medium (prepared from

987.5 ml tap water, 12.5 ml of a 10 % solution of unused cutting fluid, 0.3 g $K_2HPO_4$ and 3.5 g $NaNO_3$). In addition all the bacteria were inoculated into one flask containing the mixture of nutrient broth and cutting fluid. It was argued that this might encourage synergy between the different types of bacteria. These flasks were incubated with orbital shaking at ~ 100 rpm for 2 weeks at ambient temperature. It was anticipated that these growth conditions would allow the bacteria to develop the biochemical activities needed to metabolise cutting fluid.

The proprietary blend of micro-organisms marketed under the trade name EcoSafe HD by BioIndustries, Dublin, and a sample of the aerobic microbial waste water treatment plant operated by Lubrizol were obtained. A sample (20 ml) of each culture was inoculated into 100 ml medium (prepared from 0.3 g unused cutting fluid, 0.027 g $K_2HPO_4$ and 0.17 g $NaNO_3$ in 100 ml of tap water) contained in a 1 litre conical flask. The media were incubated with orbital shaking at about 100 rpm for 3 months at ambient temperature.

The inoculum for the bioreactor (Run 9) was prepared by mixing 50 ml aliquots of: the cultures of the individual bacteria; the culture of the defined mixture of NCIMB bacteria; the culture from EcoSafe HD, and the undefined mixed culture from Lubrizol. Part of this combined culture was taken for analysis of TOC and COD. The remainder (480 ml) was added to the bioreactor.

## Pre-Treatment of Used Cutting Fluid with Hydrogen Peroxide (Run 12)

It has been reported that pre-treatment with hydrogen peroxide ($H_2O_2$) can increase the biodegradability of certain compounds (3,4,5). To test the effect of $H_2O_2$, the pH of 4 litres of used cutting fluid (nominally 10%) was adjusted from pH 7.25 to 3.7 with 30% sulphuric acid (about 10 mls). Ferrous sulphate (4.8 g $FeSO_4.7H_2O$) was added with stirring followed by 72 g of 30 % $H_2O_2$. After stirring for 100 minutes at ambient temperature, the pH of the mixture was brought back to neutrality. Approximately 1/40th of the volume was removed for analysis and the remainder was washed into the bioreactor.

## Monitoring Biodegradation

The following parameters were monitored during the trials at AWE:

- carbon dioxide output,
- concentration of oxygen dissolved in the bioreactor liquid,
- concentration of Total Organic Carbon (TOC),
- concentration of Total Inorganic Carbon (TIC),
- concentration of Chemical Oxygen Demand (COD), and
- concentration of Total Phenols.

TOC, TIC, COD and Total Phenols were measured using Dr Lange Test kits according to the manufacturer's instructions with a Lasa 20 photometer. TOC was measured using test kits LCK 381 and LCK 380 (ranges 60 -735 and 2 - 65 mg/l carbon respectively). COD was measured using test kits LCK 014, LCK 114, LCK 314 and LCK 414 (ranges 1,000 - 10,000, 150 - 1000, 15 - 150 and 5 - 60 mg/l oxygen demand respectively). Samples for phenol were measured using test kit LCK 345 (range 0.05 - 5.0 mg/l) after dilution in water.

Bioreactor samples were centrifuged at 8000 rpm in a Jouan centrifuge (model BB VVV), when the samples contained a significant amount of biomass and associated material (generally > 0.1 g/litre). This yielded a precipitate fraction of biomass plus associated material and a supernatant fraction of "solubilised" material and oil. The centrifuged pellet was resuspended in an equal volume of demineralised water and re-centrifuged. The supernatant liquid from the washed biomass was added to the original supernatant fluid and the pellet was resuspended in water. The suspension of washed biomass and the combined supernatant fluid were analysed separately, following dilution in water as required.

Samples of cutting fluid were analysed before and after biodegradation by Gas Chromatography-Mass Spectroscopy (GC-MS), or by GC with a flame ionisation detector (GC-FID). Samples of untreated cutting fluid were prepared for analysis by separating the oil by centrifugation. For GC-MS, the oil was dissolved in dichloromethane (DCM) prior to injection (1 µl), via a hot (320 °C) split (30:1) injector onto a DB-5MS capillary GC column (20m x 0.25 mm id x 0.25 µm film). The column was heated from 40 °C to 325 °C at 8 °C per minute. The output was scanned from 25 to 550 amu by a Perkin-Elmer TurboMass spectrometer at 2 scans per second. The following internal standards were used in the quantitative analyses: heptamethylnonane, chloro-octadecane, squalene, d8-naphthalene, d10-phenanthrene and d10-pyrene. For GC-FID analysis, a DCM extract was injected (1 µl) via a hot (320 °C) injector onto a DB-1 capillary GC column (30 m x 0.32 mm id x 0.25 µm film). The column was heated from 40 °C to 325 °C at 8 °C per minute. Samples of biotreated cutting fluid were analysed by GC-FID and GC-MS without centrifugation to remove oil or biomass. GC-MS was performed on a DCM extract before and after acidification. The acidic compounds extracted from the acidified sample were derivitised with N-methyl, N-trimethylsilyl, trifluoroacetamide (MSTFA) prior to GC-MS as above.

**Ultrafiltration and Activated Charcoal Filtration**

The output from the bioreactor (typically about 75 litres) was filtered through a Membrex ESP 50 portable ultrafiltration unit equipped with a 0.1 micron polyacrylonitrile membrane with a hydrophilic coating. This produced about 73 litres of permeate and 2 litres of retentate, which contained

concentrated biomass plus precipitated material. The permeate from the ultrafilter was passed through activated charcoal.

## Solidification of Biomass plus Associated Material

The feasibility of immobilising biomass plus associated material in cement for disposal in the UK's national repository for LLW was checked using a slurry prepared by ultrafiltration of a batch of biotreated fluid. The slurry contained about 3% w/v dry weight biomass and associated material. The concentration of the solids in part of the slurry was increased to about 7.5% dry weight by adding the deposits (containing ~ 20% dry weight solids) that had been scraped off the side of the bioreactor at the end of several runs. After autoclaving the slurries at 121°C for 15 minutes, about 500 g was mixed with about 1000 g Ordinary Portland Cement using a Kenwood food mixer. The cement matrices were dispensed as 100 g and 500 g amounts into 50 ml and 250 ml plastic bottles respectively. The bottles were capped and placed in vermiculite so that the cement could set in an insulating environment. After 7 days, the plastic bottle was cut away from the cement and the 500 g sample was examined for compressive strength in an Instron Compressive Load Cell / Test Machine. An initial load of 400 $kN/m^2$ was applied for 15 minutes. There was no evidence of any cracking during this period, so the load was uniformly increased until failure occurred. The 100 g samples were resuspended in water (volume in ml equal to 20 times the surface area of the cement sample in $cm^2$). The visual appearance of the leachate water was recorded after 7 or 28 days and the TOC was measured using Dr Lange test kits. The leachate water was also extracted with 1,1,2-Trichloro-1,2,2-Trifluoroethane to enable the content of Total Petroleum Hydrocarbon to be measured using FTIR spectrometry.

## RESULTS

The studies at AWE were initiated using a microbial culture selected from waste cutting fluid that had been stored on site for some years. It was argued that the indigenous population would have adapted to metabolise the various components of the cutting fluid. The culture system selected for development at AWE was based on batch growth in a stirred tank bioreactor (1). An advantage of the stirred tank was that it allowed representative samples to be taken.

The extent of biodegradation was followed throughout the run by measuring the amount of carbon in 3 fractions:

1) carbon that was released as carbon dioxide.
2) carbon that remained either soluble or emulsified in the supernatant liquid upon centrifugation of bioreactor samples.
3) carbon that precipitated upon centrifugation. This carbon was derived from the biomass and from any insoluble material that precipitated with the biomass.

When biodegradation studies at AWE were initiated, the first three runs in the bioreactor converted only a small amount of organic material to carbon dioxide (1) (Table III). Attempts were made to increase the degree of biodegradation by changing the bioreactor conditions (1). The following changes were evaluated:

- use of various old stocks of cutting fluid (which were presumed to contain well-established microbial populations),
- use of tap water instead of demineralised water, and use of a lower initial concentration of cutting fluid (Run 4 onwards, except Run 10),
- not applying mechanical stirring to the bioreactor (Runs 7 and 8),
- using a different microbial source (Runs 9 and 10),
- pre-oxidation of the cutting fluid with hydrogen peroxide (Run 12).

Table III. Carbon Dioxide Production Per Unit of COD Initially in the Bioreactor

| Run | Conditions | $CO_2$ / COD (g per mg/l x 1000) |
|---|---|---|
| 1 | Indigenous Microbes , ~ 0.5 % Cutting Fluid, Bioreactor Stirred, Demineralised Water | 2.9 |
| 2 | Indigenous Microbes , ~ 2 % Cutting Fluid, Bioreactor Stirred, Demineralised Water | nd |
| 3 | Indigenous Microbes , Cutting Fluid from various old stocks, Bioreactor Stirred, Demineralised water | 2.6 |
| 4 | Indigenous Microbes , ~ 0.1 % Cutting Fluid, Bioreactor Stirred, Tap water | 8.1 |
| 5 | Indigenous Microbes , ~ 0.1 % Cutting Fluid Added to Run 4, Bioreactor Stirred, Tap water | 10 |
| 6 | Indigenous Microbes , ~ 0.3 % Cutting Fluid, Bioreactor Stirred, Tap water | 6.6 (8.6 after 54 days) |
| 7 | Indigenous Microbes , ~ 0.1 % Cutting Fluid, Bioreactor Not Stirred, Tap water | 7.5 |
| 8 | Indigenous Microbes , ~ 0.1 % Cutting Fluid, Bioreactor Not Stirred, Tap water | 7.3 |
| 9 | Non-Indigenous Microbes From Commercial Sources, ~ 0.3 % Cutting Fluid, Bioreactor Stirred, Tap water | 6.5 (14 days) |
| 10 | Non-Indigenous Microbes From Commercial Sources,, ~ 2 % Cutting Fluid, Bioreactor Stirred, Tap water | nd |
| 11 | Back to Indigenous Microbes, ~ 0.3 % Cutting Fluid, Bioreactor Stirred, Tap water | 5.7 |
| 12 | Indigenous Microbes, $H_2O_2$ Pre-treatment of ~ 0.3% Cutting Fluid, Bioreactor Stirred, Tap water | 9.3 |

*nd not determined because first peak of $CO_2$ production was off-scale.*

The effect that the various treatments had on mineralisation was assessed by comparing the amount of carbon dioxide produced per unit of initial Chemical Oxygen Demand (COD) (Table III). The initial COD reflected the concentration of cutting fluid in the bioreactor at the start of the run. The conclusions from this comparison were:

- Carbon dioxide production per unit of initial COD markedly increased after Run 3, when the bioreactor conditions were changed by reducing the initial concentration of cutting fluid and by using tap water instead of demineralised water (1).
- Carbon dioxide production was not stimulated by not stirring (Runs 7 and 8).
- The use of a commercial blend of microbes did not stimulate biodegradation (although they were examined for only 14 days under the more favourable conditions in Run 9).
- Pre-treatment with hydrogen peroxide (Run 12) stimulated biodegradation to a slight extent. Although the amount of carbon dioxide produced per unit of initial COD in Run 12 was greater than that in Runs 6 and 11, it was not greater than that achieved in Run 5.
- Carbon dioxide production per unit of initial COD was in fact greatest for Run 5. This Run was started by adding a low concentration of cutting fluid at the end of a previous run (1).

When the efficacy of these treatments was assessed in terms of the amount of carbon dioxide produced per unit of COD added (Table III) it appeared that Runs 5 and 12 produced about 50 % more carbon dioxide than Runs 6 and 11. However, when these treatments were assessed in terms of the distribution of carbon between the 3 fractions, carbon dioxide, "soluble / emulsified carbon" and biomass plus associated material (Table IV), Runs 5 and 12 were not markedly different to the others.

It was not until the bioreactor conditions were varied in Runs 4, 5 and 6 that there was a measurable degree of biodegradation (1) and this occurred in 2 main peaks of activity, as shown by carbon dioxide production and oxygen consumption. A similar pattern of activity was found in subsequent runs. A typical pattern is shown in Figure 1a for Run 11. This activity was accompanied by a fall in the amount of solubilised / emulsified organic material (Figure 1b) as indicated by the reduction of the COD.

Biodegradation was not complete. The added organic carbon was converted in approximately equal proportions to carbon dioxide, organic carbon in solution/ emulsion, and biomass plus any associated insoluble material (see data from Runs 4, 5, 6 and 11 in Table IV.

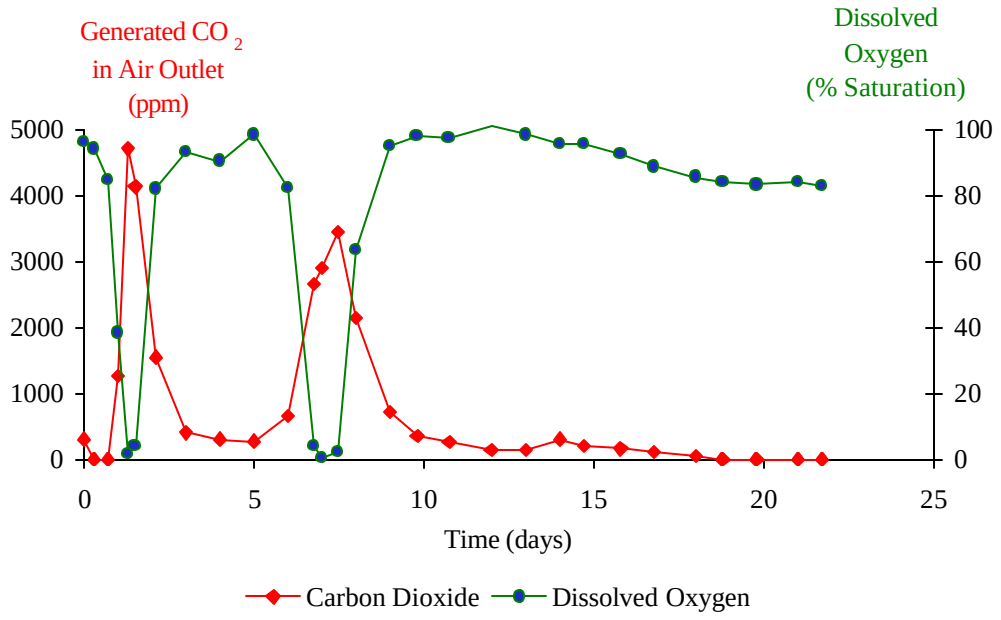


Fig. 1a. Pattern of Biodegradation of ~ 0.3 % Waste Cutting Fluid in Run 11

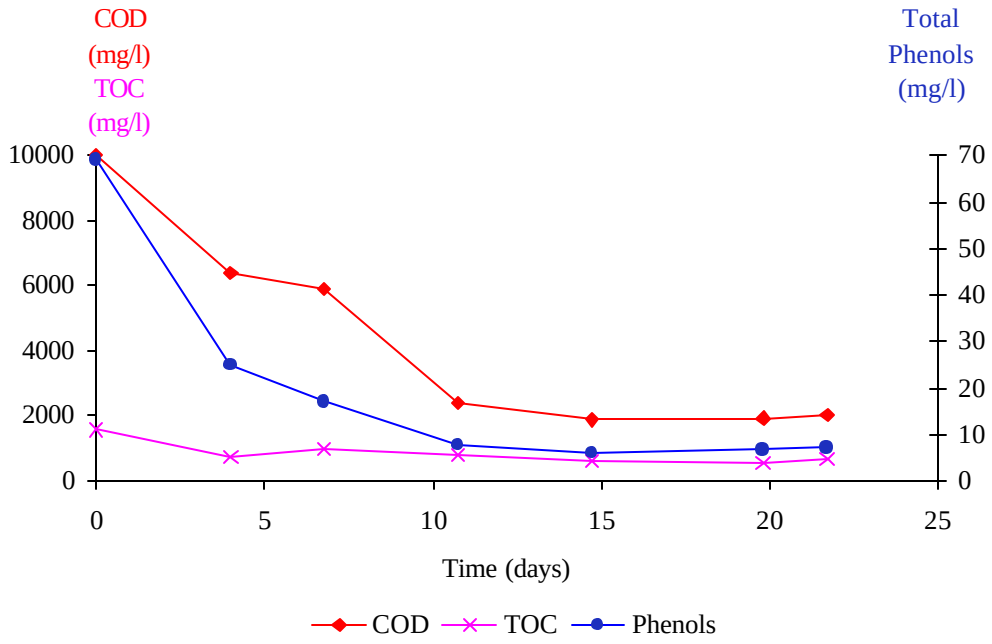


Fig. 1b. Removal of Organic Material During Biodegradation of ~ 0.3 % Waste Cutting Fluid in Run 11.

Table IV. Distribution of Organic Carbon

| Run | Distribution * of Organic Carbon (%) | | |
|---|---|---|---|
| | Carbon Dioxide | "Soluble" Carbon | Biomass + Associated Material |
| 4 | 45 | 28 | 27 |
| 5 | 38 | 31 | 31 |
| 6<br>(54 days) | 38<br>(45) | 33<br>(29) | 28<br>(26) |
| 9 | 36 | 31 | 33 |
| 11 | 36 | 33 | 31 |
| 12 | 38 | 22 | 40 |

* *Distribution calculated from the amount of carbon measured in carbon dioxide, biomass + associated precipitated material and in solution/emulsion (at ~ 21 days unless stated otherwise).*

The lack of complete mineralisation was not due to inhibition by the biocide. For example the concentration of phenols in Run 11 fell by about 90 % (Figure 1b). Likewise, it was not due to incomplete degradation of the oil. The concentration of hydrocarbon before and after biotreatment in Run 11 was measured using GC-FID. At the start of the run the concentration of hydrocarbon in the bioreactor fluid was 2130 mg/litre. After biodegradation it had fallen to 150 mg/l, which represents 93 % biodegradation.

It seemed likely that the residual organic material was mainly one or more product of the partial biodegradation of the hydrocarbon in the waste cutting fluid. Published information on the biochemical pathways for hydrocarbon degradation suggested that these products could be carboxylic acids (6, 7, 8, 9, 10). Analysis by GC-MS of a methylene chloride extract of acidified cutting fluid before and after biodegradation in Run 11 indicated that the biodegraded sample contained organic acids that were not present in the waste cutting fluid prior to biodegradation. The concentration of these acids (650 mg/l) would have made a significant contribution to the TOC measured at the end of the biodegradation run (total TOC of "soluble/emulsified" fraction and biomass plus associated material = 1310 mg/l). There

was some evidence that these acids belonged to a group of saturated higher fatty acids derived from the cyclic alkanes (or naphthenes) that are present in crude oil. The cyclic alkane fraction of crude oil is considered to be less readily biodegradable in the natural environment than straight chain alkanes and simple aromatics (8, 11).

The performance shown by Run 11 was typical and so it was decided to use the degree of biodegradation shown by Run 11 to evaluate the efficiency of a waste disposal process based on biodegradation. It was considered that the overall waste disposal process would involve biodegradation followed by downstream processing of the biotreated fluid.

## Downstream Processing of the By-Products from Biodegradation

It was envisaged that downstream processing would involve ultrafiltration of the biotreated fluid followed by adsorption onto activated charcoal. It was reported previously that this would produce a liquid that would qualify for processing in the radioactive aqueous waste treatment plant at AWE (1). Ultrafiltration would remove biomass and associated insoluble material from the biotreated fluid. It was envisaged that some biomass would be returned to the bioreactor and that the surplus would be immobilised in cement for disposal as solid radioactive waste in the UK's repository for low level radioactive waste that is managed by British Nuclear Fuels Ltd. at Drigg.

Accordingly, the feasibility of immobilising biomass in cement was examined. A biomass slurry was concentrated by ultrafiltration to about 3% dry solids, which was the limit with the available equipment. The concentration of solids in the UF retentate slurry was increased by adding to part of the retentate the deposits (containing about 20% dry weight solids) that had been scraped off the side of the bioreactor at the end of several runs. These 2 biomass slurries were then incorporated into cement in such a way that the slurries provided all the water in the cement matrices. The cement matrices contained either about 1% or about 3 % biomass plus associated material (Table V).

The disposal to Drigg of cement matrices prepared from liquids containing oils depends on certain conditions being met (12). The 2 main conditions are that the matrix should withstand a compressive pressure greater than 0.4 $N/mm^2$ and that oil should not leach from the matrix when it is immersed in water for 7 and 28 days; i.e. oil should not be visible on the surface of the leach water. Both cement matrices prepared from the biomass fraction had a compressive strength that was 10 x greater than that required and both did not visibly release oil upon immersion in water (Table V). A small amount of organic and inorganic material was leached from the test samples and an unidentified white precipitate was formed. Nevertheless, the leach waters contained <50 µg/l total petroleum hydrocarbons and <10 mg/l total organic carbon. Thus, it should be possible to incorporate excess biomass (and any associated material) into cement for disposal as solid waste.

Table V. Cementation of Biomass - Compressive Strength and Leachability

| **Concentration of Biomass plus Associated** Material in Matrix (dry wt / wt matrix) (%) | Composition of Cement Matrix | | Ultimate Compressive Strength of Matrix ($N/mm^2$) | Appearance of Leach Water | Total Petroleum Hydrocarbons in Leach Water (µg/litre) | | Total Organic Carbon in Leach Water (mg/litre) | |
|---|---|---|---|---|---|---|---|---|
| | Ordinary Portland Cement (grams) | UF Retentate (grams) | | | After 7 days | After 28 days | After 7 days | After 28 days |
| 1 % | 1067 | 533 | 12.0 / 13.1 | White precipitate, but no oil | < 50 / < 50 | < 50 / <50 | nd / ~ 2 | 4.1 / 6.8 |
| 3 % | 1109 | 554 | 6.9 / 7.3 | White precipitate, but no oil | < 50 / < 50 | < 50 / < 50 | 8.6 / 7.3 | 2.4 / nd |

Values shown for duplicate samples

nd = not detected

## DISCUSSION

### Viability of the Waste Disposal Process

Biodegradation would form part of a multi-stage process for the disposal of radioactive cutting fluid (Figure 2).

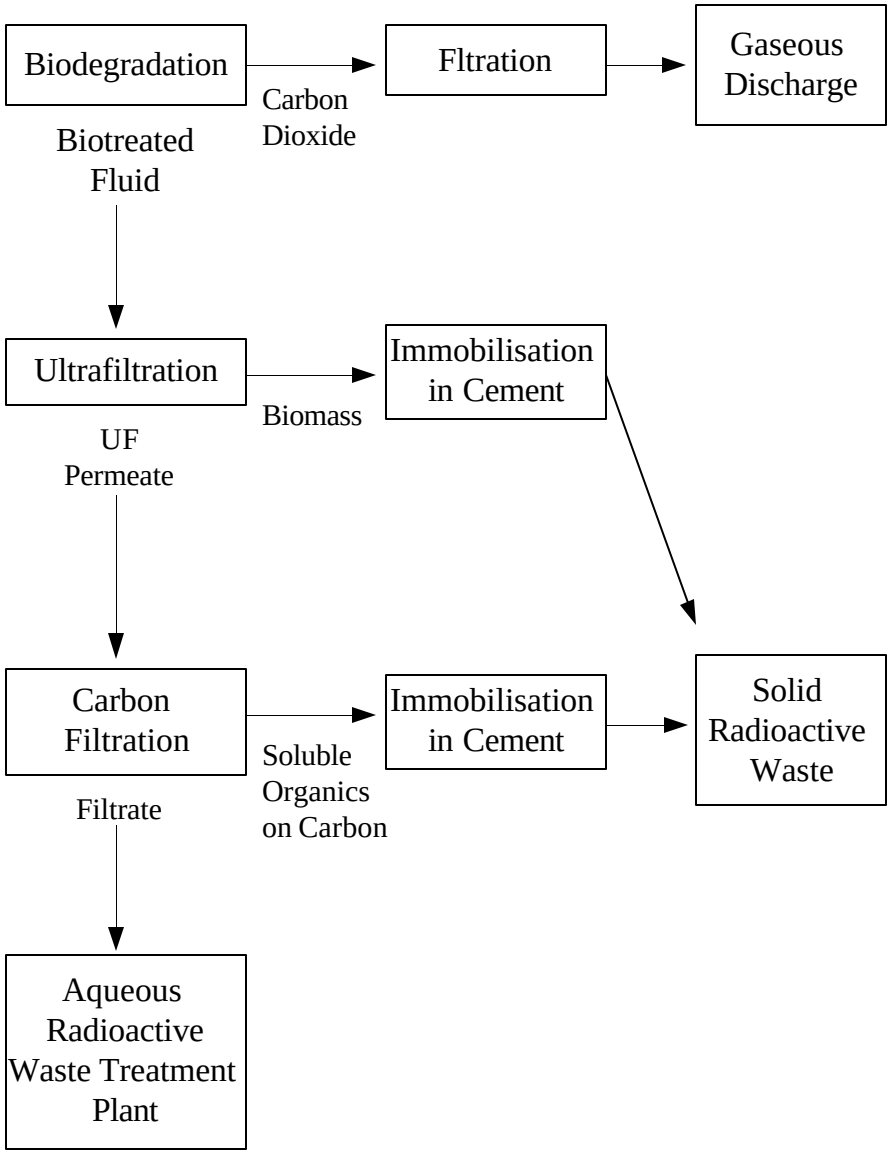


Fig. 2. Proposed Treatment Process for Waste Radioactive Cutting Fluid

The viability of this waste disposal process has been assessed by estimating the amount of secondary waste arising from the biodegradation of 1000 kg of waste cutting fluid. It was assumed that the cutting fluid was used at a 10 % w/w dilution of a formulation containing 77% w/w carbon. An approximately equal distribution of organic carbon between the 3 fractions: carbon dioxide, soluble / emulsified compounds, and biomass plus associated material (Table III) would lead to the formation of:

- 93 kg carbon dioxide,
- 39 kg of soluble / emulsified organics, if it assumed that these are represented by cyclohexane carboxylic acid,
- 51 kg biomass plus associated material, if it is assumed that all this material has a similar composition to biomass and contains 50 % carbon by weight (6).

Carbon dioxide would be vented to the atmosphere. This would generate a relatively small amount of extra secondary waste, i.e. HEPA filters and pipework.

It was reported at WM'99 that the soluble / emulsified material could be adsorbed onto activated charcoal (1). Assuming that activated charcoal could adsorb 20 % of its weight as organic material, and that the loaded charcoal could be immobilised in cement up to 30 % w/w of the final cement matrix, the disposal of 39 kg of "soluble / emulsified" material would generate 647 kg of solid waste.

The present study has shown that "biomass plus associated material" can be incorporated into cement (Table V), although it was not possible to examine a biomass loading greater than 3 % of the matrix. At this loading the formation of 51 kg of this material would lead to the production of 1,700 kg of cement waste.

## CONCLUSION

The biodegradation process under the conditions outlined above would produce about 2,300 kg of secondary cement waste per 1000 kg of waste cutting fluid processed (at a nominal 10% dilution of the formulated product). This compares with an estimated 2000 kg of solid waste per 1000 kg of waste cutting fluid that would generated by absorbing the cutting fluid on clay granules prior to immobilisation in cement. In addition, biodegradation would involve 4 unit processes: bioreactor treatment, ultrafiltration, activated carbon filtration and cementation. Additional processing entails additional equipment that would have to be disposed of as radioactive waste at the end of the project. Thus, at present, it appears that a waste disposal process based on biodegradation would be more inefficient than direct cement solidification of the untreated waste cutting fluid.

However, a number of ways of improving the efficiency of the biodegradation process can be envisaged. For example, it should be possible to reduce the amount of biomass for disposal by recycling it to the bioreactor. In addition, it should be possible to improve bioreactor performance by stimulating

the degree of biodegradation of the residual organic material. The studies at AWE did not systematically examined the effect of a wider range of different growth conditions, such as mineral nutrients, vitamins and other organic growth factors or temperature. Neither did it examine other bioreactor systems.

Furthermore, there are potential opportunities to reduce the amount of solid waste arising from the by-products of biodegradation. There is scope to increase the loading of biomass in cement and to either optimise the chosen techniques for downstream processing or select more suitable techniques. Thus, it is concluded that biodegradation of waste radioactive cutting fluid still has the potential to be developed into a viable process.

## REFERENCES

1. G.T. Taylor, N.D. Knee, M.J.H. Keep and V.C.M. Freestone, "Biodegradation of Used Machine Tool Cutting Fluids", In: Proceedings of Waste Management 99, Tucson, USA, February 28 - March 4, 1999, WM Symposia Inc. (1999).
2. M.J.H. Keep, "Management and Disposal of Radio-active Machine Tool Coolants at AWE Aldermaston", In: Proceedings of Waste Management 98, Tucson, USA, March 1 - 5, 1998 WM Symposia Inc. (1998).
3 A R Bowers, S H Cho and Singh A, "Chemical Oxidation of Aromatic Compounds; Comparison of $H_2O_2$, $KMnO_4$ and $O_3$ for Toxicity Reduction and Improvements in Biodegradability" in Proceedings of the First International Symposium, Chemical Oxidation Technology: Technology for the Nineties, Vanderbilt University Nashville, USA, February 1991, Edited by W.W Eckenfelder, A.R Bower. and J.A Roth, Technomic Publishing Co.
4 W Eul, G Scherer and O Helming, "Practical Applications of Hydrogen Peroxide for Wastewater Treatment" 1991 in Proceedings of the First International Symposium, Chemical Oxidation Technology: Technology for the Nineties, Vanderbilt University Nashville, USA, February 1991, Edited by W.W Eckenfelder, A.R Bower. and J.A Roth, Technomic Publishing Co.
5. R C Linneman and T H Flippin, "Hydrogen Peroxide Pre-treatment of Inhibitory Wastestream - Bench-scale Treatability Testing to Full Scale Implementation: A Case History" in Proceedings of the First International Symposium, Chemical Oxidation Technology: Technology for the Nineties, Vanderbilt University Nashville, USA, February 1991, Edited by W.W Eckenfelder, A.R Bower. and J.A Roth, Technomic Publishing Co.
6. M. Alexander, "Biodegradation and Bioremediation". Academic Press, San Diego, USA. (1994). ISBN 0-12-049860-X.
7. C.E. Cerniglia, "Microbial Transformation of Aromatic Hydrocarbons". In: Atlas R.M. (Ed.), Petroleum Microbiology. Macmillan Publishing Co., New York, USA, pp. 99-128, (1984). ISBN 0-02-949000-6.
8. J.J. Perry, "Microbial Metabolism of Cyclic Alkanes". In: Atlas R.M. (Ed.), Petroleum Microbiology. Macmillan Publishing Co., New York, USA, pp. 61-97 (1984). ISBN 0-02-949000-6.

9. P. Pitter and J. Chudoba, "Biodegradability of Organic Substances in the Aquatic Environment", CRC Press, Boca Raton, USA (1990). ISBN 0-8493-5131-6.
10. E.M Singer and W.R. Finnerty, "Microbial Metabolism of Straight-chain and Branched Alkanes". In: Atlas R.M. (Ed.), Petroleum Microbiology. Macmillan Publishing Co., New York, USA, pp. 1-59 (1984). ISBN 0-02-949000-6.
11 A.C. Van der Linden, "Degradation of Oil in the Marine Environment". In: Watkinson, R.J. (Ed.), Developments in Biodegradation of Hydrocarbons - 1. Applied Science Publishers Ltd., Barking, UK, pp. 165-200 (1978). ISBN 0 85334 751 4.
12 K.G. Elgie, P. Grimwood and A. Coyle, "Drigg Guidance Note: Conditioning of Wastes by Solidification". BNFL Waste Management and Decommissioning, LLW Management Services. Reference: DGN01, Issues 3, December 1996.

## FOOTNOTES:

[a] AWE (formerly known as the Atomic Weapons Establishment) is responsible for the design, manufacture, in-service support and eventual retirement of the UK's nuclear deterrent. It is managed as a government owned, contractor operated organisation by a joint venture company created by BNFL, Lockheed Martin and Serco.