PROPOSAL FOR MANAGEMENT OF BIOMEDICAL MIXED WASTES

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ABSTRACT

Mixed radioactive and chemical wastes generated by biomedical research were characterized and various treatment methods evaluated. These wastes consist primarily of organic solvents used in the extraction and purification of radiolabelled biomolecules that are contaminated with low levels of the long-lived radionuclides, ^{3}H and ^{14}C, and fall into three broad solvent categories: phenol/chloroform, acetonitrile/water, and mixtures of miscellaneous solvents, such as benzene, carbon tetrachloride, and other hazardous chemicals. Currently, there is no commercial disposal outlet for mixed wastes nor may they be stored on-site legally for more than 90-180 days. Most of these mixed wastes can be effectively rendered into nonradioactive chemical and radioactive aqueous wastes, both of which can be disposed of legally. However, treatment of mixed waste requires an expensive and difficult-to-obtain Resource Conservation and Recovery Act (RCRA) Part B permit for licensure as a treatment, storage, and disposal facility. More significantly, such treatment is expensive and presents potential occupational hazards from the direct handling of waste materials. Deregulation of certain mixed wastes by the Nuclear Regulatory Commission would be the safest, most cost-effective, and practical method for dealing with much of the biomedical mixed wastes. In any event, it is important that a national regulatory solution be found.

INTRODUCTION

In response to the 1985 EPA rule banning land burial of hazardous chemical wastes, the commercial low-level radioactive waste (LLRW) sites, all of which utilize shallow land burial, refused to accept contaminated organic chemical wastes. While shallow land burial is the only disposal technology in use for solid LLRW in the U.S., the EPA specifically forbids the burial of most hazardous chemical wastes. As a result, no legal disposal outlet for mixed waste exists. By default, generators have been forced to store this waste on site, only legal for periods ranging from 90-180 days.

At The Rockefeller University, mixed wastes consist of spent organic solvents that have been contaminated with ^{3}H- and ^{14}C-labelled biomolecules and their precursors, e.g., amino acids, peptides, nucleotides, intermediates, and metabolites. Although these wastes contribute less than 1% of the radioactivity to our total LLRW inventory, they present a compliance problem since there is no disposal outlet and treatment of mixed wastes entails a complicated permitting procedure. We have categorized the mixed wastes produced at our institution, their chemical and radioactive composition, and the laboratory procedures which generate them. A variety of simple bench-scale separation and treatment protocols were also evaluated.

Over the past five years, The Rockefeller University has accumulated 38 individual containers of mixed waste, ranging in volume from 100 mL to 4 L each, for a total volume of approximately 75 L. Several are multi-phasic mixtures and, in most cases, were labelled by their generators as "unknown" with regard to isotope, activity, and chemical composition. Figure 1 provides an overview of our work.

Mixed wastes were analyzed for their radioactive composition by beta and gamma counting and their chemical composition by gas chromatography (GC) and gas chromatography/mass spectrometry (GCMS). Only the isotopes ^{3}H and ^{14}C were detected, although it is not possible to rule out ^{32}P, ^{35}S, ^{125}I, ^{51}Cr, or other short-lived radioisotopes which would have decayed to background levels by the time of analysis. Several individual items contained considerable levels of radioactivity, e.g., 370 - 925 kBq mL^{-1} (10 - 25 μCi mL^{-1}), but most were only slightly contaminated, e.g., <3.7 kBq mL^{-1} (<0.1 μCi mL^{-1}). Mixed wastes with only background levels of radioactivity were disposed of as chemical waste without any further study. Table I summarizes the characterizations.

Chemical analyses showed that the mixed waste fell into three broad categories: phenol/chloroform mixtures, acetonitrile/water mixtures, and a third group of miscellaneous solvents including the carcinogens benzene and carbon tetrachloride.

Phenol/chloroform waste consists of two phases of approximately equal volume: an upper, aqueous phase of water, methanol, and phenol (approximately 60%, 30%, and 10%, respectively); and a lower, organic phase of chloroform, phenol, and methanol (approximately 60%, 25%, and 15%, respectively). In molecular biology, phenol, chloroform, and 1:1 mixtures of these chemicals are routinely added to aqueous solutions to extract nucleic acids and denature proteins that remain after enzymatic digestions. The aqueous and organic phases were separated in a separatory funnel. Phenol in the aqueous phase was removed by chromatography through a column of synthetic resin (XAD-4 Styrene-Divinylbenzene Copolymer Resin, Rohm and Haas) yielding a radioactive eluent of water and a small

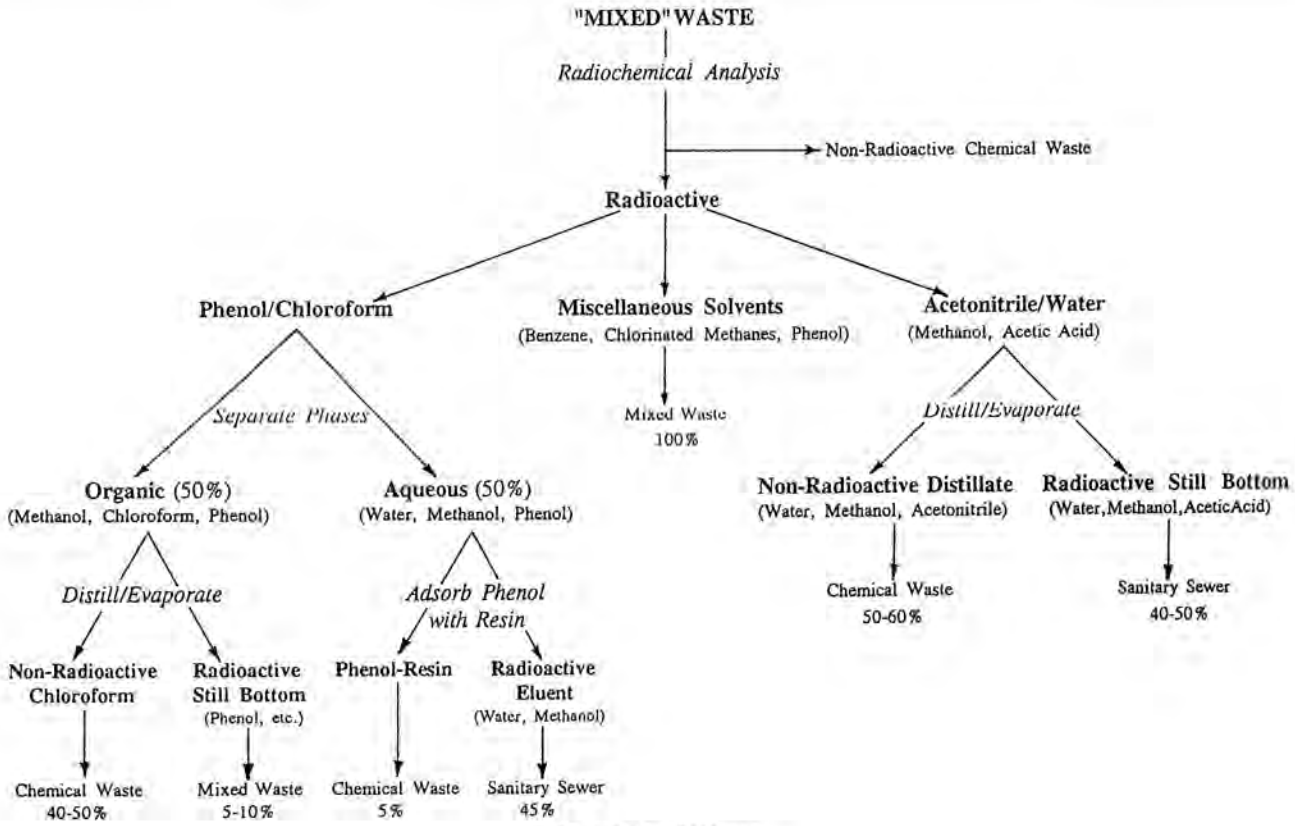

Fig. 1. Mixed Waste.

TABLE I

Mixed Wastes Generated by Biomedical Research

		Radioactive Concentrations, kBq mL^{-1}						
		^{3}H			^{14}C			
Group	Typical Components	Mean	SD	Median	Mean	SD	Median	Volume (L)
Phenol/ Chloroform	Chloroform, Phenol, Methanol, Water	158.73	331.52	4.07	0.41	0.08	0.41	18.00
Aqueous Acetonitrile	Acetonitrile, Water, Methanol, Acetic Acid, Ethanol	10.36	17.02	4.44	1.29	17.02	0.48	30.65
Miscellaneous Solvents	Chlorinated Methanes, Phenol, Methanol, Benzene, Toluene-and Pseudocumene-based Scintillation fluors	19.98	39.59	2.33	0.29	0.41	0.11	24.70
Total		50.01	101.80	3.64	0.74	7.27	0.34	73.35

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The acetonitrile/water mixtures principally contain acetonitrile and water but methanol, acetic acid, and a trace of dimethylformamide are also present. These wastes are generated during the purification of radiolabelled proteins and lipids by high performance liquid chromatography (HPLC). Distillation and flash evaporation yielded a nonradioactive distillate of acetonitrile, water, and methanol, all of which can be disposed of as aqueous chemical waste. The still bottom, in which the radioactivity concentrated, consisted of water and low concentrations of methanol and acetic acid and is suitable for disposal as aqueous radioactive waste.

The third class of mixed waste, the complex group of miscellaneous solvents, appears to be the result of the consolidation of smaller volumes of chemically-compatible waste generated during routine organic chemistry. Our principal source is a laboratory that synthesizes large amounts of tritiated steroids, generating, among other solvents, radioactively-contaminated benzene and carbon tetrachloride. Due to the inherent toxicity of these materials, this group of mixed waste was left in storage.

Over the past two decades, laboratory protocols for biomedical and basic biological research have been progressively scaled down from the liter to milliliter to microliter scale, reducing both reagent costs and the volume and activity of wastes. Radioactively-contaminated solvents are now generated slowly and our inventory of mixed waste has increased only slightly over the last few years. Advances in enzyme-catalyzed *in vitro* procedures (e.g., polymerase-catalyzed reactions, reverse transcriptase, nick translation), the availability of high specific activity precursors, and a larger variety of commercially-available radiolabelled biomolecules have helped supplant many of the less efficient *in vivo* techniques. The improving sensitivity of many nonradioactive assays may further minimize mixed waste generation. Nonetheless, the use of radioactivity is essential to biomedical research and thus mixed wastes will remain a necessary by-product of this work.

Our work suggests that mixed wastes from biomedical research can be effectively managed by a series of simple, routine, laboratory procedures. Unfortunately, they are labor-intensive, cost-ineffective, and increase the likelihood of occupational exposures to both radiation and toxic organic chemicals. Since the levels of radioactivity are, on average, very low, and the volumes are very low, we believe that deregulation by the NRC would allow their disposal as hazardous chemicals in the most efficient and environmentally and occupationally appropriate method. The NRC deregulation of liquid scintillation fluids in 1981 facilitated the disposal of the major category of biomedically generated LLRW. By declaring the various organic chemicals contaminated with low levels of long-lived radioisotopes below regulatory concern, "mixed waste" would, for the most part, be eliminated from biomedical institutions.