A TECHNIQUE FOR SEPARATION OF XENOBIOTICS FROM LARGE AMOUNTS OF LIPIDS

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Organic pollutants with a general distribution throughout our environment can be detected in air, water, sediment and biota. The highest concentrations of xenobiotics in the environment are found in animals at high throphic levels such as in predator fish, birds and mammals. It is in these animal species we can first expect to see health effects due to the pollutants. The majority of xenobiotics determined in the environment so far are substituted with halogen atoms. It is however unknown how many more compounds with or without halogen substituents that may be present in the environment and to what extent these substances are hazardous to life. It is thus of importance that the analytical technique permit as low detection limits as possible in order to identify and quantify as many xenobiotics as possible. Furthermore, it is also of interest to isolate anthropogenic compounds from biological material for toxicological studies and bioassays. In both cases it is a prerequsite that endogenous lipids have been removed from the samples. It is most common that the techniques used for removal of lipids restrict the amounts of material that can be used as starting material in the work up procedures. Both the use of alkali or strong acid, KOH and H_2SO_4, respectively, are used for removal of lipids but these methods are destructive to labile componds in the matrix[1]. Partititioning procedures[2] and gel permeation chromatography (GPC)[3] can be used without destruction of labile substances but still the total amounts of lipids that can be removed are limited. In the present study a method for removal of large amounts of lipid from fish and seal samples and isolation of xenobiotics in a few fractions is reported.

Substances in herring oil (1500 g) has been partioned between the oil and acetonitril in a cyclic procedure. The partitioning was checked by use of ^{14}C-labelled hexachlorobenzene (HCB) that is known to have a poor partitioning coefficient between oil and acetonitril and thus is a good indicator compound for development and evaluation of the method. In order to transfer 50% of DDE in herring oil (1 part) it is necessary to use 10 times that volume of acetonitrile - this is by definition 1 batch. The cyclic partitioning procedure was adjusted so that the herring oil was treated with one batch acetonitrile per day (15 l). After 5 batches (75 l of acetonitrile) as much as 96.6% of the HCB was transfered to the acetonitrile fraction. The lipid weight was simultanously reduced to 19.5% of the original weight. When the procedure was repeated on the residual lipids but in a smaller scale, ca. 300 g lipids was treated with 3 batches of acetonitrile (9 l), 98.3% of the HCB was recovered in the acetonitrile together with 7.8% of the lipids (117 g). HCB together with other neutral compounds, dissolved in hexane, were isolated in the neutral hexane phase after treatment with KOH (0.5 M). Acidic compounds were retreived in hexane after acidification of the alkaline phase. The residual lipids in the neutral fraction

was 30 g and in the alkaline phase ca. 90 g.

The details of the separation of lipids from xenobiotics will be reported. Furthermore, the separation of the residual neutral compounds from the lipids (2%) and also fractionation of the acidic compounds will be shown. The partitioning coefficients between oil and acetonitrile of several other radiolabelled xenobiotics and their metabolites, such as individual PCB congeners and their hydroxylated and methyl sulphonyl metabolites, pentachloropenol, chlorinated paraffins and polychloroguaiacols, has been determined.

The method has so far proven to be highly efficient in the reduction of large amounts of lipids from fish oil. The potency of the method may permit the isolation of large quantities of xenobiotics from environmental samples for biological testing without the interference of lipids. Since these large amounts of lipids are removed the concentration factor is increased more than two orders of magnitude. This will permit potential detection of xenobiotics present in the samples at lower concentrations than earlier reported.

Acknowledgement

Financial support was given by the Swedish Environmental Protection Agency.

References

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