MEASUREMENTS OF POLYCYCLIC AROMATIC HYDROCARBON (PAH) BIOACCESSIBILITY AND THEIR USE IN THE ASSESSMENT OF HUMAN HEALTH RISK

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Both road traffic and the combustion of domestic fuels are considered by many to be the main sources of polycyclic aromatic hydrocarbons (PAHs) in urban and industrial areas. A recent pollution survey of UK soil and herbage indicated approximately 5-7 times greater concentrations of benzo(a)pyrene in urban and industrial soils than in rural soils. The incorporation of bioaccessibility data is a valuable tool in the assessment of human health risk in regard to polycyclic aromatic hydrocarbon soil contamination.

This study has shown that the RIVM method is a suitable technique for the bioaccessibility testing of polycyclic aromatic hydrocarbons. The bioaccessibility results obtained for all three soils in this study were in general found to agree with previously published values as well as predicted values based on relative fugacities. These predicted bioaccessibilities were calculated using published relative fugacities and the measured benzo(a)pyrene bioaccessibility of a spiked standard soil. Fugacity was shown to be a good indicator of bioaccessibility relative to benzo(a)pyrene.

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INTRODUCTION

Polycyclic aromatic hydrocarbons are a group of chemicals that are formed during the incomplete combustion of organic substances (e.g. coal, oil, gas, wood, rubbish, etc). They are found throughout the environment in the air (attached to dust particles), water, and soil. There are more than 100 different polycyclic aromatic hydrocarbons, which generally occur as complex mixtures not as single compounds, and the health effects of individual polycyclic aromatic hydrocarbons differ. With respect to human health risk, the 16 polycyclic aromatic hydrocarbons typically analysed for are: acenaphthene (ACE), acenaphthylene (ACY), anthracene (ANTH), benzo(a)anthracene (BaA), benzo(a)pyrene (BaP), benzo(b)fluoranthene (BbFLN), benzo(g,h,i)perylene (BghiP), Benzo(k)fluoranthene (BkFN), chrysene (CHR), dibenzo(a,h)anthracene (DBahA), fluoranthene (FLN), fluorene (FLR), indeno(1,2,3-cd)pyrene (IPY), naphthalene (NPH), phenathrene (PHN) and pyrene (PYR). These are often referred to as the USEPA priority polycyclic aromatic hydrocarbons. Under current UK waste regulations this list is extended to include coronene.

Generally more is understood about these 16 polycyclic aromatic hydrocarbons and there is a greater chance that you will be exposed to them. They are thought to be the more harmful of the polycyclic aromatic hydrocarbons and exhibit harmful effects that are representative of polycyclic aromatic hydrocarbons as a group.

ASSESSMENT OF POLYCYCLIC AROMATIC HYDROCARBONS

Current Environment Agency guidance regarding the "UK approach for evaluating human health risks from petroleum hydrocarbons in soils" (Environment Agency, 2005) on non-threshold compounds is to analyse for individual indicator compounds and to apply Soil Guideline Values / Generic Assessment Criteria to these compounds without considering additive effects of either the remaining fractions or other indicator compounds. There are currently no published Soil Guideline Values available for polycyclic aromatic hydrocarbons. Generic Assessment Criteria have been published by Nathanial *et al.* (2007) for benzo(a)pyrene, fluorene, napthalene and di-benzo(a,h)anthracene and were calculated using the CLEA UK (beta) model version 1.0. This model has been used to calculate Generic Assessment Criteria for the remaining polycyclic aromatic hydrocarbons.

Known additive affects between different priority carcinogenic polycyclic aromatic hydrocarbons have been modelled by Brown *et al.* (1999) and a similar hazard index approach to considering polycyclic aromatic hydrocarbons has been adopted. The individual hazard quotients are calculated using the measured concentration (or the level of detection as lower bound concentrations) of each substance divided by the relevant assessment criteria. These are then summated to provide the hazard index. The risk is considered acceptable if the hazard index is <1.

Polycyclic aromatic hydrocarbon concentrations in the majority of urban areas in the UK exceed the published Generic Assessment Criteria (Nathanial *et al.* 2007). The latest DEFRA suggestions regarding risk assessment methodology in the UK (DEFRA, 2006) addresses this situation by considering adopting an index dose based on an excess life-time cancer risk of 10^{-4} under Part 2A. This same approach could be adopted for determining contamination under the planning policy PPS 23. However, DEFRA (2006) appears to suggest that the burden of proof under planning will be required to be much more stringent. It is considered that this level of excess risk is

consistent with the concept of the "significant possibility of significant harm", as opposed to the level of excess on which the Soil Guideline Values / Generic Assessment Criteria derivation is based as stated in DEFRA CLAN 2/05 (DEFRA, 2005).

In the assessment of risk, the use of bioaccessibility testing can be a useful approach in assessing harm as part of a detailed site-specific risk assessment and in the derivation of site-specific assessment criteria.

Assessment of bioaccessibility

Bioaccessibility testing is a method of assessing bioavailability. The definitions of bioaccessibility and bioavailability are: (1) absolute bioavailability – absorbed dose / administered dose (x 100), (2) relative bioavailability – absorbed fraction from soil / absorbed fraction from dosing medium used in toxicity study (x 100), (3) bioaccessibility – amount extracted from soil during test / total concentration in soil (x100).

Recent studies by Pu *et al.* (2004) have indicated that the Physiological Based Extraction Test (PBET) is a useful method of assessing polycyclic aromatic hydrocarbon bioavailability in soils, and provides reproducible results which can be correlated with *in-vivo* rat study data. The measured polycyclic aromatic hydrocarbon bioaccessibilities in this study were suitably adjusted to consider variability and method uncertainty, and have been used in a risk assessment as a surrogate for relative bioavailability. It should be noted that tests that provide a measure of bioavailability should be treated with caution as intestinal metabolism of polycyclic aromatic hydrocarbons can increase transfer rates through the epithelium (Cavret and Feidt, 2005) and any test method would need to ensure that this had been taken into account. The use of bioaccessibility measurements as a surrogate for bioavailability assumes 100% transfer through the epithelium.

The method adopted for polycyclic aromatic hydrocarbon bioaccessibility was the RIVM method developed by Oomen *et al.* (2001) as shown diagrammatically in Figure 1. This is one of a number of different PBET tests all of which are designed to mimic to a greater or lesser extent transit through the human digestive system. The method uses three digestion media to simulate digestion in the mouth, stomach and intestine. The test is conducted at a controlled temperature of 37ºC and is summarised as follows: (1) add 0.6 g of prepared soil sample (dry weight) to 9 ml saliva and incubate for 5 minutes; (2) add 13.5 ml gastric juice and incubate for 2 hours; (3) add 27 ml duodenal and pancreatic juices with 9 ml bile, and incubate for 2 hours; and (4) centrifuge for 5 minutes and analyse the extract. The method described by Oomen *et al.* (2001) was carried out with one modification; agitation was carried out using a rotary shaker in a temperature controlled water bath rather than an end-over-end agitator in a temperature controlled cupboard.

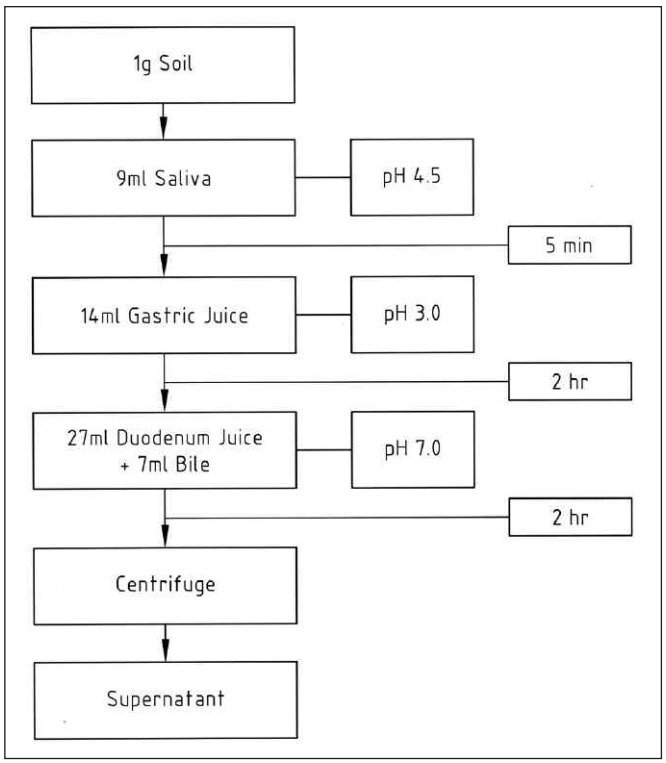

Figure 1. *Diagrammatic procedure of RIVM method (after Pu* et al.*, 2004).*

The RIVM method differs from the more commonly used Ruby or modified Ruby methods (Ruby *et al.*, 1996; Rodriguez *et al.*, 1999) in that the RIVM method also adopts an oral phase prior to the normal stomach and intestinal phases. Extracts are not removed sequentially in the RIVM method and so information regarding bioaccessibility at each digestion phase is not obtained, i.e. it only provides a measure of total bioaccessibility. As such, it is not valid for contaminants that are known to precipitate within the intestine e.g. cationic metals (British Standards, 2007).

The artificial digestion media are also significantly more complex in the RIVM method requiring greater laboratory precision and technique to prepare. The solutions are also less stable and cannot be stored. At the time of undertaking the test, no established UK soils laboratory offered the RIVM method as a standard test.

Validation

The method was validated for polycyclic aromatic hydrocarbon bioaccessibility using standard artificial soil spiked with benzo(a)pyrene (OECD, 1984). Results were compared with published values of bioaccessibility for such a spiked soil. The results of the validation testing on the spiked soil provide good correlation with published estimates of benzo(a)pyrene bioaccessibility and also provide good evidence for repeatability of the test method.

Results

The results of bioaccessibility testing for three different soils are shown on Figure 2. Results are compared with the predicted relative bioaccessibility for polycyclic aromatic hydrocarbons, which were based on the relative fugacities of individual polycyclic aromatic hydrocarbons to benzo(a)pyrene (Brown *et al.*, 1999). Relative fugacities used are shown in Table 1. Predicted relative bioaccessibilities were calculated using the relative fugacity and the measured benzo(a)pyrene bioaccessibility.

It is evident that the measured bioaccessibility and predicted bioaccessibility show a good degree of correlation, albeit conservative, with the exception of di-benzo(a,h)anthracene (where two of the soils exhibited higher bioaccessibilities than that predicted). On the whole, fugacity was a good indicator of bioaccessibility relative to benzo(a)pyrene. The measured bioaccessibility of benzo(a)pyrene in all three soils did not exceed 20%.

The results indicate that the measured bioaccessibility of polycyclic aromatic hydrocarbons fall broadly into two groups. The first group has a relatively low bioaccessibility of less than 30% and the second group displays a higher bioaccessibility in the range of 60% to 85%. The compounds in the second group were acenaphthene; acenaphthylene; anthracene; dibenzo(a,h)anthracene; fluorene; naphthalene and phenathrene.

A study by Tang *et al.* (2006), which carried out polycyclic aromatic hydrocarbon bioaccessibility analysis using the Ruby method (Ruby *et al.*, 1996) also indicated two broad groupings, with acenaphthene; acenaphthylene and naphthalene having an oral bioaccessibility of 50% or greater in both the gastric and small intestine phases, and significantly lower bioaccessibilities for most of the other compounds.

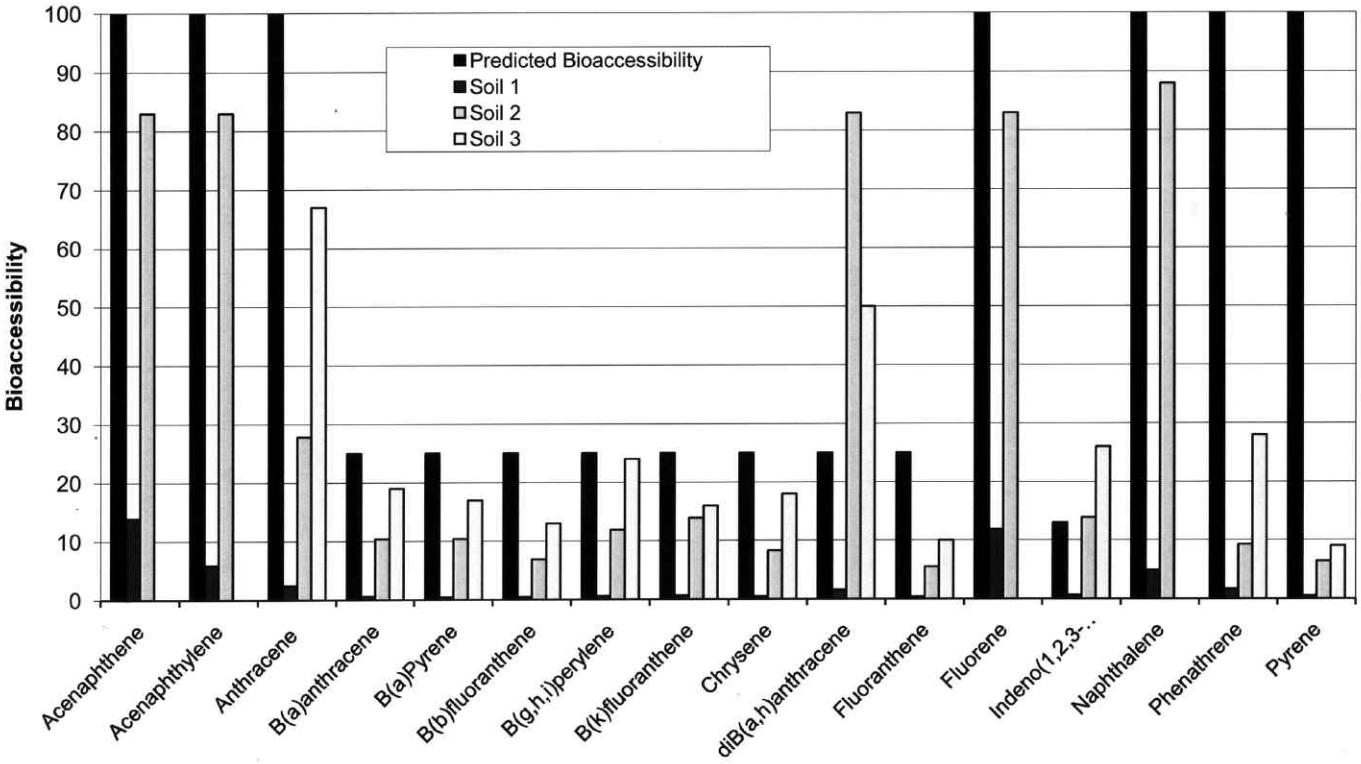

Figure 2. *Predicted and measured bioaccessibility values for the 16 priority polycyclic aromatic hydrocarbons.*

Compound	EC no.	C fraction	Fugicity	Relative fugicity
Acenaphthene	15.5	C12-C16		4.00
Acenaphthylene	15.06	C12-C16		4.00
Anthracene	19.43	C16-21		4.00
Benzo(a)anthracene	26.37	C21-C28		1.00
Benzo(a)Pyrene	31.34	C28-C35	1.00	1.00
Benzo(b)fluoranthene	30.14	C28-C35		1.00
Benzo(g,h,i)perylene	34.01	C28-C35		1.00
Benzo(k)fluoranthene	30.14	C28-C35		1.00
Chrysene	27.41	C21-C28		1.00
diBenzo(a,h)anthracene	33.92	C28-C35		1.00
Fluoranthene	21.85	C21-C28		4.00
Fluorene	16.55	C16-C21		4.00
Indeno(1,2,3-cd)pyrene	35.01	>C35		0.52
Naphthalene	11.69	C10-C21		4.00
Phenathrene	19.36	C16-C21		4.00
Pyrene	20.8	C16-C21		4.00

Table 1. *Relative fugacity compared to benzo(a)pyrene.*

Other studies by Hack and Selenka (1996) and Oomen *et al.* (2000) measured total polycyclic aromatic hydrocarbon bioaccessibilities ranging between 10% and 60%. These were significantly higher than the measured bioaccessibilities by Van de Wiele (2004) who reported the release of 0.1 – 1.4%. These differences were ascribed by Van de Wiele (2004) as possibly being a result of their study being on historically contaminated soils, which could show aging effects.

To demonstrate the potential usefulness of incorporating polycyclic aromatic hydrocarbon bioaccessibilities, a risk assessment was carried out using mean UK urban soil data (Environment Agency, 2007) assessed against generic assessment criteria derived using the CLEA UK (beta) model version 1.0. Hazard quotients were calculated for both a residential with garden land-use and a residential without garden land-use (i.e. no home-grown plant uptake). Figure 3 compares the hazard quotients for the 16 polycyclic aromatic hydrocarbons without taking any bioaccessibility data into account. It is evident without the incorporation of bioaccessibility that in typical UK urban soils, benzo(a)anthracene, chrysene and pyrene will exceed their assessment criteria, which implies that average UK residential gardens have potentially unacceptable concentrations of these three hydrocarbons. Figure 3 also demonstrates the relative importance of the ingestion of home-grown produce. For benzo(a)anthracene and chrysene, contamination via plant uptake is a significant pathway. Whereas for benzo(a)pyrene, di-benzo(a,h,)anthracene and pyrene, plant uptake is a less significant pathway.

Incorporation of bioaccessibility data into the derivation of site-specific assessment criteria can be used within polycyclic aromatic hydrocarbon risk assessments. The effects on the hazard quotients are shown in Figure 4, indicating that the incorporation of such data has a major effect on all of the above polycyclic aromatic hydrocarbons. When considering the combined pathways of soil ingestion and the consumption of home-grown produce, the hazard quotient for benzo(a)anthracene, benzo(a)pyrene and chrysene can be reduced by some 40 – 60%. For chrysene and benzo(a)pyrene, consideration of default soil to plant concentration factors is also necessary before the risk of these compounds can be considered acceptable.

Conclusions

The study has shown that the RIVM method is capable of producing consistent and repeatable results when applied to spiked standard soils. The method has been shown to provide reasonable correlation with *in vivo* rat studies and *in vivo* test results and can be considered a suitable method for the bioaccessibility testing of polycyclic aromatic hydrocarbons. The bioaccessibility results obtained for all three soils in this study were in general found to agree with previously predicted values, which were based on relative fugacities. The use of fugacity as a predication for polycyclic aromatic hydrocarbon bioaccessibility has been shown to be useful.

Incorporation of bioaccessibility data can be valuable in the assessment of risk to human health posed by polycyclic aromatic hydrocarbons in urban UK soils. A reduction in risk of between 40% - 60% has been shown for benzo(a)anthracene,

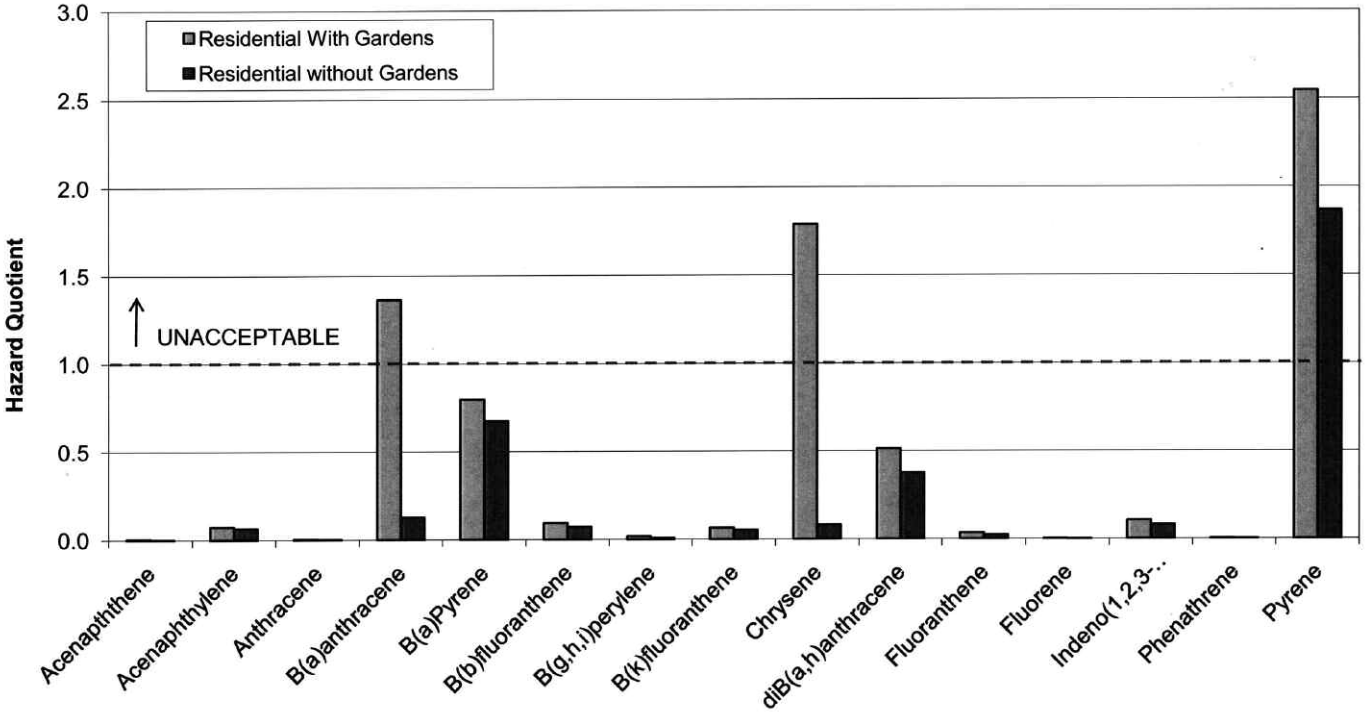

Figure 3. *Calculated hazard quotients for mean urban soil concentration.*

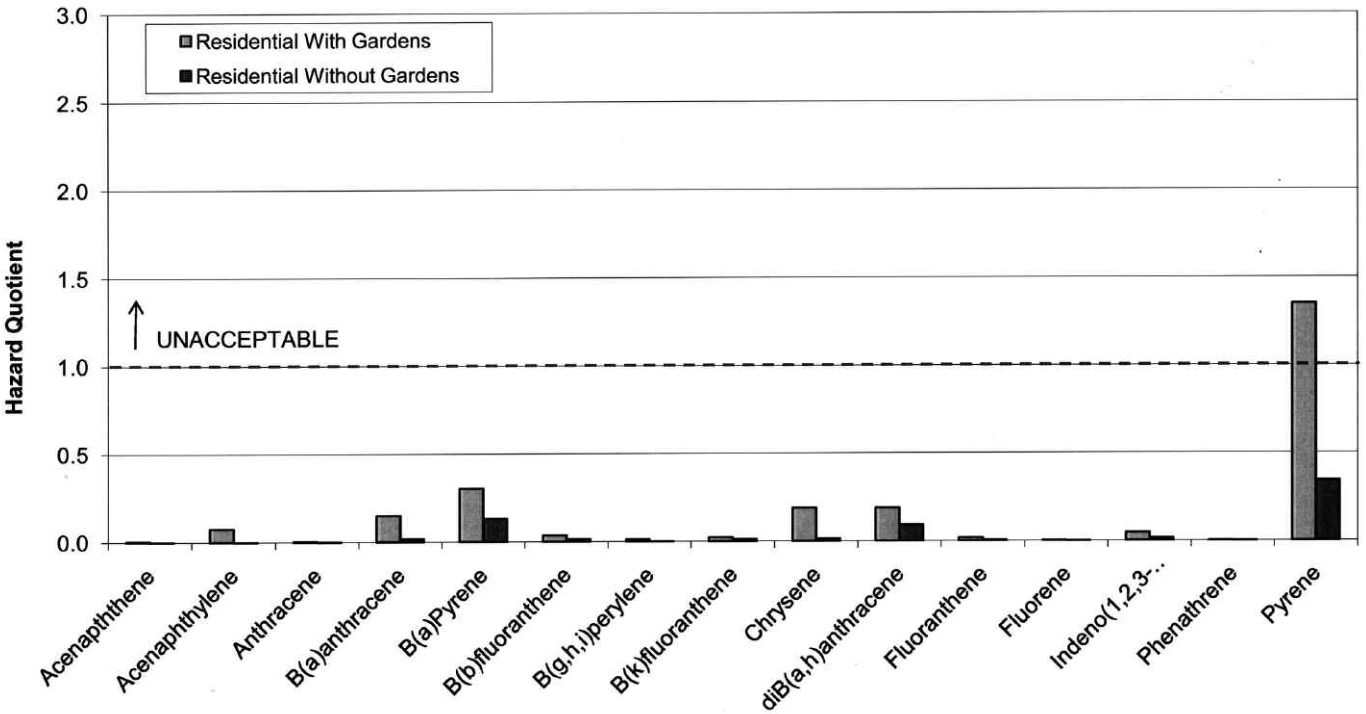

Figure 4. *Calculated hazard quotients for mean urban soil concentration including bioaccessibility results and including revised soil to plant concentration factors for benzo(a)anthracene and chrysene.*

benzo(a)pyrene and chrysene when incorporating bioaccessibility data. The incorporation of such bioaccessibility data can be used to provide justification for levels of polycyclic aromatic hydrocarbon contamination commonly found in UK urban soils. It is considered that use of bioaccessibility testing and data as a risk assessment tool should be progressed for polycyclic aromatic hydrocarbon contaminated soils.

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