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Biodegradation and bioaccumulation rate of PCBs in cows

Alaa Kareem Mohammed
Asst. Prof.
Genetic Engineering and Biotechnology
Institute for Postgraduate Studies
Baghdad University, Baghdad, Iraq

Abstract

The object of this research is to focus on bioaccumulation and biodegradation rate of hydrophobic, persistent pollutants namely PCBs in cows. A simple mathematical mass balance equation which represents the input and output streams of these persistent pollutants to a cow was developed. Samples were taken from rural agricultural farms in Abu-Greab in Baghdad. These samples were analyzed for input and output terms which represent vegetation, soil, concentrate, milk and feces to measure the concentration of PCBs in these samples. It was found that bioaccumulation and biodegradation rate of PCBs in cows is a bout 202.6 µg/d. and most of PCBs input the cow is by vegetation rather than concentrate or soil while PCBs input to the cow by air or water are very low values and can be neglected so as the PCBs out put the cow by urine and air. Also it was found that the output PCBs from the cow is about 62.5% by milk and 37.5% by feces.

Keywords: PCBs, Hydrophobic contaminants, Pesticides. Contaminants in cows

معدل تحلل وتراكم المبيدات (PCBs) في البقر

الخلاصة:

يهدف هذا البحث الى دراسة معدل تحلل وتراكم المواد العضوية صعبة التحلل نوع (PCBs) التي تدخل اجسام البقر في المزارع الريفية القريبة من مناطق التلوث بهذا النوع من المواد . وضعت معادلة موازنة المادة والتي تم فيها حساب معدل دخول وخروج هذه الملوثات في اجسام البقر. اخذت نماذج عديدة من منطقة ريفية زراعية في منطقة ابي غريب في بغداد. تم تحليل النماذج التي اخذت من العلف والتربة والتقاوي والحليب وبراز البقر. لقد وجد ان معدل تحلل وتراكم المواد نوع (PCBs) تبلغ 202.6 مايكروغرام / اليوم. وان هذه المواد تدخل البقر عن طريق الطعام (العلف) اكثر منه عن طريق التقاوي و التربة . ان تركيز الملوثات الداخلة الى البقر عن طرق الماء والهواء يكون قليل جدا ويمكن اهماله وكذلك تركيز الملوثات في البول قليل ويمكن اهماله. كذلك وجد ان 62.5% من هذه الملوثات تخرج جسم البقرة عن طريق الحليب بينما 37.5 % يخرج عن طريق البراز.

1- Introduction

Many chemicals formed industrially such as organic chemicals are released to air, water, and soil and enter humans primarily through food in particular through meat and dairy products [1,2]. One of these compounds is a polychlorinated biphenyl (PCB) which consists of 2 to 10 chlorine atoms attached to biphenyl. The chemical formula for a PCB is $C_{12}H_{10-x}Cl_x$. PCBs are very stable compounds and do not decompose readily. This is due to their chemical inability to oxidize and reduce in the natural environment [3]. Furthermore, PCBs have a long half life (8 to 10 years) and are insoluble in water, which contributes to their stability. Their destruction by chemical, thermal, and biochemical processes is extremely difficult, and presents the risk of generating extremely toxic dibenzodioxins and dibenzofurans through partial oxidation [4].

Cow's milk and dairy products are a major source of human exposure to persistent, lipophilic compounds. In general, they contribute 27% of human exposure to polychlorinated biphenyls (PCBs). Because of their bioaccumulatory properties, the transfer of (PCBs) through some food chains has been quite thoroughly studied [5]. Dairy cattle take up these chemicals via feed, primarily grass. Atmospheric deposition is usually the primary vector of lipophilic chemicals into grass. However, cattle do ingest soil and if the soil contamination is high compared to atmospheric levels then the soil can be a significant source of chemicals like PCBs in cow's milk, and thereby an important vector of human exposure[6]. The goal of this study was to establish and verify a mass – balance equation model for the evaluation the rate of bioaccumulation and biodegradation of lipophilic organic chemicals like PCBs in the cows.

2- Materials and methods

Experimental work includes detection the concentration of PCBs in samples represented the vegetation, soil, concentrate, milk and feces to verify the mathematical mass balance equation mentioned above. This can be achieved by conducting the following steps:-

2-1. Lyophilization:

Water must be eliminated from samples (which could be soil, vegetation, foods, milk, or feces), because it will interfere with chromatographic analysis. Lyophilization procedure allows removing water by sublimation without losing pollutants. Samples are first freezed in Lyophilization chamber at (-40) ^oC and vacuum of 1 mbar for ½ hr. Finally the sample is heated to 5 ^oC and water is removed by sublimation in a 24 hr.

Fig.(1) Lyophilization chamber.

2-2. Extraction:

Pollutants and organic matter have to be extracted from the matrices and put into a solution with polar solvents. This is done by the soxhlet apparatus. 150 gm of sample is put into a cellulose extraction thimble (material which is porous enough to let the solvent flow and retain the matrix particles). 100 ml of a mixture of hexane and acetone 1:1 is used as a solvent. The mixture is heated by the plate and evaporates from the flask, and through the bypass sidearm will reach the condenser tube. By condensing, it will drip on the thimble and the sample slowly filling the extraction tube and so slowly extracting pollutants and organic matter from the sample. When solvent level inside the extraction tube reaches the level of the apex of reflux sidearm, it will flow back to the flask. This cycle will be repeated many times and complete extraction (with recovery of 90 – 95 %) will be completed in a 12 hr time. Usually for every three sample a blank is run in parallel to check eventual contamination due to extraction procedures and atmospheric dusts. At the end of extraction in flasks we have a solution of pollutants and organic matter dissolved in solvents.

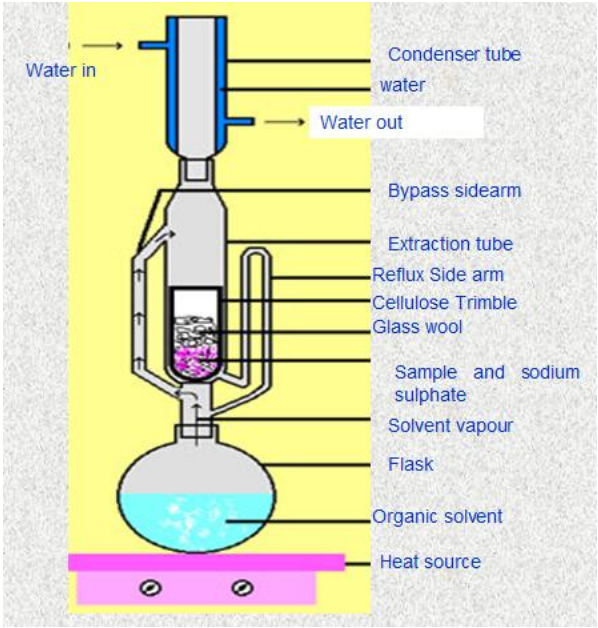


Fig. (2) Soxhlet apparatus used for extraction process.

2-3. Evaporation:

The 100 ml of solvent used for extraction must be concentrated to 3 ml using rotary evaporator. The flask containing the solvent is heated by a water bath to 40^{o}C. Evaporated solvent is driven away from the flask by low vacuum, condensed and distilled away to another flask. Care is needed to avid complete drying of the sample.

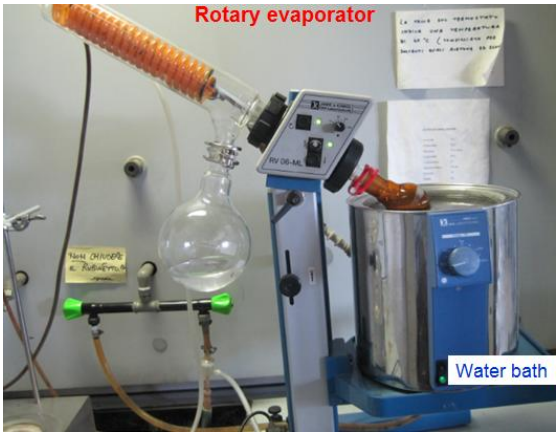
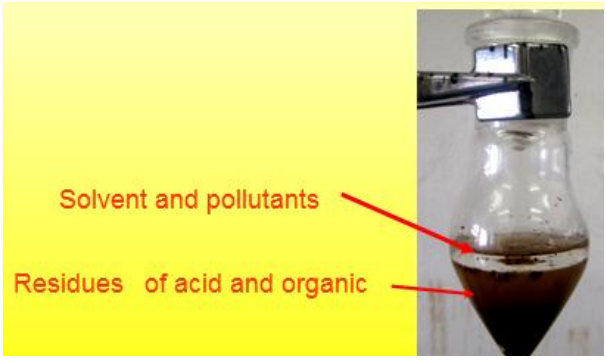


Fig. (3) Rotary evaporator used for evaporation.

2- 4. Acid digestion:

After being reduced to 3 ml, samples are transferred to small, heart shaped flask, 6 ml pure H2SO4 is added directly inside the flask. Then the flask is stoppered and sealed with parafilm. Acid digestion is an overnight procedure. Sulphuric acid digests organic matter but not PCBs since these pollutants are stable and acid resistant. Then two phase solution is formed. The lower phase consists of residues of acid and burned organic matter (dark brown colour). The upper phase consists of solvent and pollutants (now free from organic matter. The upper phase (3ml) is separated and transferred to another hearted shape flask and concentrated to 1-1.5 ml by rotary evaporator. The sample is transferred to 1 ml vial. Gentle nitrogen flow is used to concentrate the sample exactly to 1 ml, accelerate the evaporation and prevent the intake of moisture and dust.



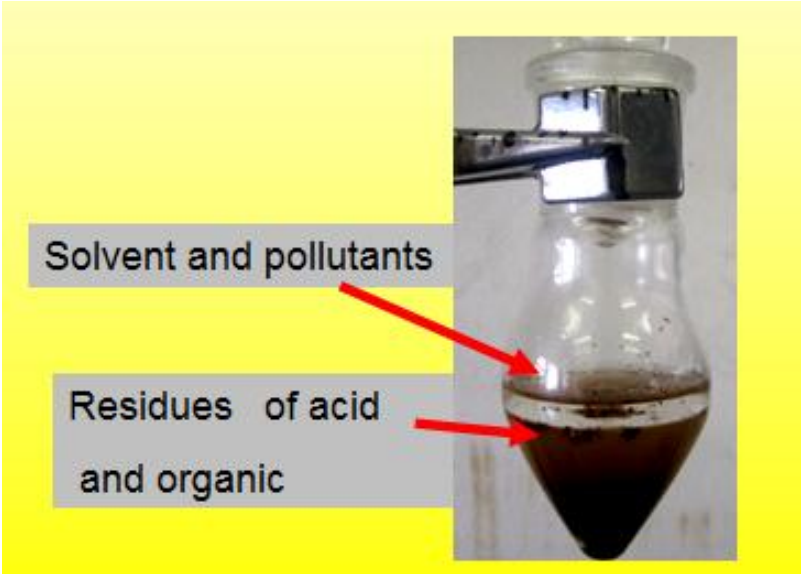


Fig.(4) Heart shaped flask used for acid digestion.

2-5. Purification by chromatographic column:

Samples are purified by a two phase chromatographic column. The column is filled with 10 gm of partially deactivated silica gel and then with10 gm of florisil (100 – 200 mesh) to adsorb the more polar organic compounds rather than pollutants. The packed column is washed with 50 ml of a solution (Hexane: Acetone: Dichloromethane 8:1:1) then the sample is put inside the column. The sample is eluted with a second solution of 50 ml of Hexane and then with a third solution 50 ml Hexane: Dichloromethane 1:1. 1 ml of iso-octane (2, 2, 4 triethilpentane) is added to prevent drying during storage of samples. The 100 ml of solvent must be concentrated (first with rotary evaporator and then with gentle nitrogen flow) to 1 ml and transferred to the vials. The samples are now ready for GC/MS analysis.

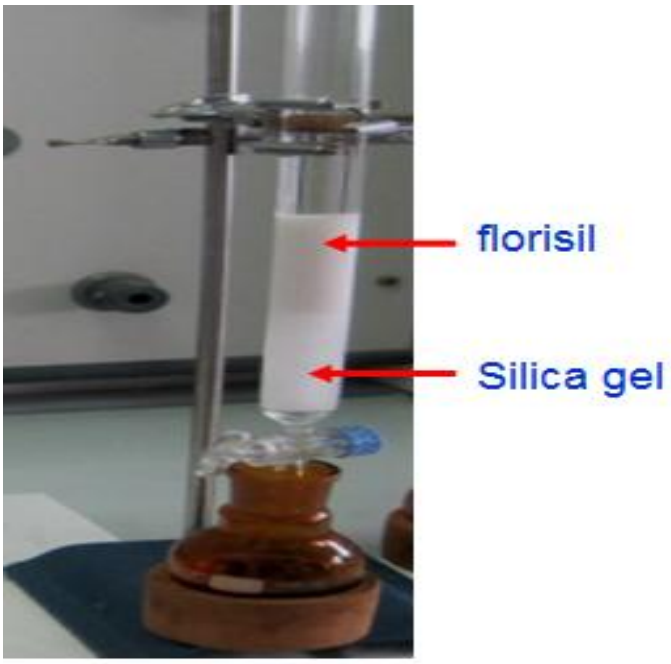


Fig.(5) Purification by chromatographic column

3- Result and discussion

3-1. Mass balance equation

The steady state mass balance equation performed very well and is simple:

Input – Output = Accumulation + biodegradation

The input streams include vegetation, soil, concentrate, air and water while out put streams includes milk, feces, urine and air.

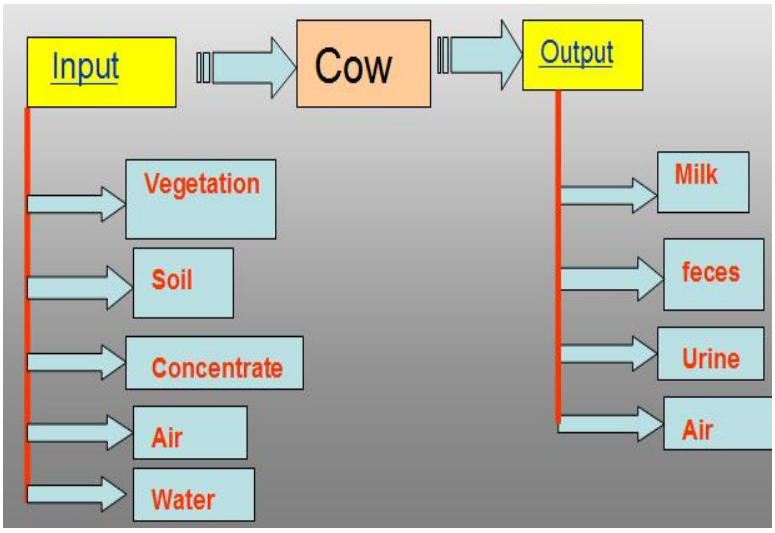


Fig. (6) Schematic diagram of input and output streams of PCBs to the cow

Analysis of samples show that the PCBs concentrations are negligible in air , water and urine. This result was in agreement with that found by Bennett [7,8].Table (1) shows the concentration of PCBs in milk, concentrate, vegetation, feces and soil

Table (1) PCBs number and concentration in different streams

PCBs No	Milk	Concentrate	Vegetation	Feces	soil
18	< LOQ	< LOQ	< LOQ	< LOQ	< LOQ
31+28	0.025	< LOQ	0.141	< LOQ	< LOQ
52	0.089	< LOQ	0.358	< LOQ	< LOQ
44	< LOQ	< LOQ	< LOQ	< LOQ	< LOQ
101	0.295	< LOQ	0.802	< LOQ	0.342
149	0.292	< LOQ	1.128	0.11	< LOQ
118	0.465	0.014	1.071	0.244	0.024
153	1.629	0.058	5.352	0.229	0.104
138	1.031	0.048	3.202	0.266	< LOQ
180	1.19	0.045	3.078	0.485	0.076
170	0.241	< LOQ	0.621	< LOQ	0.068
194	< LOQ	< LOQ	0.226	< LOQ	0.116
209	< LOQ	< LOQ	< LOQ	< LOQ	< LOQ
Total PCBs	5.257	0.165	16.078	1.334	0.73

All values are expressed as ng /g dry weight.
LOQ : Limit of Quantification

3-2. Input streams PCBs

The quantities of PCBs input to the cow can be calculated by product the quantity input times the concentration. Table (2) shows the calculations of input PCBs to the cow

Table (2). The input quantities of PCBs to the cow.

Item	Quantity (kg/d)	Concentration (ng/g dw)	PCBs (µg/d)
Vegetation	13.5	16	216
Concentrate	2	0.165	0.33
Soil	0.335	0.73	0.224
Water	-	-	-
Air	-	-	-
			Σ=216.5

3-3. Output streams PCBs

Also the quantities of PCBs output the cow can be calculated in the same way. Table (3) shows these quantities.

Table (3). The output quantities of PCBs from the cow

Item	Quantity (kg/d)	Concentration (ng/g dw)	PCBs (µg/d)
Milk	1.651	5.257	8.68
Feces	3.9	1.334	5.202
Urine	-	-	-
Air	-	-	-
			Σ=13.88

3-4. Bioaccumulation and biodegradation rate of PCBs

By substituting the calculated values of input and output into the mass balance equation, the rate of bioaccumulation and biodegradation of PCBs in the cow can be calculated as shown below:

Input – Output = bioaccumulation + biodegradation

216.5 mg/d - 13.88 mg/d = bioaccumulation + biodegradation

Hence,

Bioaccumulation + biodegradation = 202.6 µg/d

Bioaccumulation term represents a non biodegradable PCBs like PCB No. 118, 153 138,180 while biodegradation represents ready biodegradable PCBs such as PCB No. 18, 31,44,170 and 52.[9,10]. The fraction of PCBs out put by milk and feces can be calculated as follows:

$$\text{PCBs output by milk} = \frac{8.68}{13.88} x100 = 62.5\%$$

$$\text{PCBs output by feces} = \frac{5.202}{13.88} x100 = 37.5\ \%$$

This result is in agreement with that mentioned by Mc Lachlan [11] who also concluded that about 15.4% of PCBs burden in the animals is biodegradable while 84.6% is not-biodegradable so it may accumulate in the fat fraction of tissues.

4- Conclusion

1- Most of PCBs input the cow is by vegetation rather than concentrate or soil and atmospheric deposition is the primary vector of lipophilic chemical into grass and then to cow.

2- The PCBs input to the cow by air or water are very low values and can be neglected. Also the PCBs out put the cow by urine and air can be neglected.

3- The rate of bioaccumulation and biodegradation of PCBs in the cow is about 202.6 µg/d.

4-The output PCBs from the cow is about 62.5% by milk and 37.5% by feces.

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