

# Environmental Fate of Polyhexamethylene Biguanide

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**Abstract** Polyhexamethylene biguanide (PHMB) is used as a bacteriocidal agent in a variety of applications from medical devices to pools, but is highly toxic to some aquatic species. The stability of PHMB in various environmental matrices was examined. 80% of PHMB was present in fortified creek samples after 20 days, but bound immediately to soils with the exception of sandy soil. PHMB was absorbed to below detectable levels by weathered concrete within 12 h. In addition, one over the counter wound care product containing PHMB was evaluated to assess environmental leaching; detectable levels (20 µg/mL) were still present after 1 week.

**Keywords** Polyhexamethylene biguanide (PHMB) · Environmental fate

PHMB (Polyhexamethylene biguanide) is a chemical germicide used in a variety of applications: as a preservative in wet wipes, to prevent microbial contamination in wound irrigation and sterile dressings, and to disinfect medical/dental utensils and trays (Arch Chem Co 2008). However, it is estimated that approximately 95% of the PHMB used is for pool and spa water treatment (EPA 2004). Medical devices have PHMB as the active microbiocidal ingredient,

such as wound care dressings. These types of in-home medical care products are not incinerated as traditional medical waste. The increase of in-home health care provides additional opportunities for the public and the environment to come into contact with medical waste (Lichtveld et al. 1992). As over the counter products, items with PHMB are placed in regular trash and end up in landfills, or possibly on the ground or in a stream by less responsible users.

This widely used biocide has been reviewed by EPA; EPA concluded that, with the exception of occupational handlers, PHMB has a very low aggregate risk of adverse health effects to the public or environment (EPA 2004). However, some concern as to the presence of PHMB in surface waters due to discharge from swimming pools into storm drains and similar scenarios was sent to EPA in response to its conclusion that PHMB presented a low risk to the public or environment (EPA 2004). This compound is very toxic to some aquatic species (EPA 2004). With the increased detection of pharmaceuticals, hormones and other contaminants in stream water (Kolpin et al. 2002), it is possible PHMB may be present and exert some adverse effect. There is very little data in the open literature regarding the environmental fate of PHMB. For this reason, this study examined the stability of PHMB in aqueous solutions, in different soils, with weathered concrete, and leaching from medical devices.

## Materials and Methods

Sodium hydroxide, hydrochloric acid, methanol and acetonitrile were obtained from Fisher Scientific (Waltham MA, USA); triethylamine, trifluoroacetic acid, and formic acid were obtained from Sigma-Aldrich (St. Louis MO,

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USA). Phosphate buffered saline (PBS), both 1× and 10×, were purchased from Invitrogen. Distilled deionized water was produced with a Barnstead NANOpure Diamond water purification unit. Gauze containing 0.2% PHMB was obtained over the counter, while untreated gauze from the same company was purchased from the local pharmacy. Water samples were taken from two local creeks, one in Rockville, MD and the other in Columbia, MD. Soil samples used were potting soil, purchased from the local hardware store, soil dug from a backyard (Maryland red clay), and soil taken from the bottom of a local creek (sandy gravel). Aged concrete was obtained from a local construction site (FDA facilities, White Oak, Silver Spring, MD). The PHMB, trade name Cosmocil CQ (a 20% solution in water), was graciously donated by Arch Chemical Company (Norwalk CT, USA).

The concentration of PHMB was determined in aqueous solutions by measuring the absorption at 236 nm using a Beckman Coulter DU 640 spectrophotometer (Kusnetsov et al. 1997 J applied micro). The limit of quantitation for the spectrophotometer was approximately 0.1 µg/mL. PHMB was fortified at 10, 5, 2.5, 1.25 and 0 µg/mL into pure water, 1.0 N NaOH, 1× PBS, 10× PBS, 0.125–1.0 N HCl, and water taken from two local creeks. The absorption at 236 nm of these aqueous samples was measured over time (0–60 days) to assess the stability of PHMB. Aged concrete broken from a curb at the FDA White Oak Facility in Silver Spring, MD was assessed to see if PHMB could be absorbed into the concrete. Chunks about 2 cm long were broken off the large piece; 13.5 g of these chunks were placed into a 50 mL polypropylene test tube with 20 mL of either water or 10 µg/mL PHMB in water; the absorbance of these samples was read after 24 h. The remaining concrete was broken into two pieces, both with a flat area of approximately 24 cm<sup>2</sup>. In an 8 × 11 cm polypropylene tray, one large piece of concrete was placed flat side down in 100 mL of water, and one placed flat side down in 100 mL of 10 µg/mL PHMB. The absorbance of the aqueous solution was read hourly for 7 h. Soil and gauze samples were analyzed using a high performance liquid chromatographic system (Lucas et al. 2009).

The evaluation of PHMB in soil was initially assessed by placing approximately 1 g of soil in a polypropylene test tube with 10 mL water containing 50 µg/mL PHMB in water. Each of the three types of soil was immediately vortexed, filtered, extracted and run using the HPLC system (Lucas et al. 2009). Limits of detection (0.2 µg/mL) and recovery (107%) of aqueous solutions using this HPLC system has been reported (Lucas et al. 2009). There was no PHMB detected in either the clay or the potting soil. Subsequent soil experiments used only the sandy soil. Triplicate samples of the sandy soil were placed in a polypropylene test tube with 10 mL water containing 100 µg/mL PHMB and

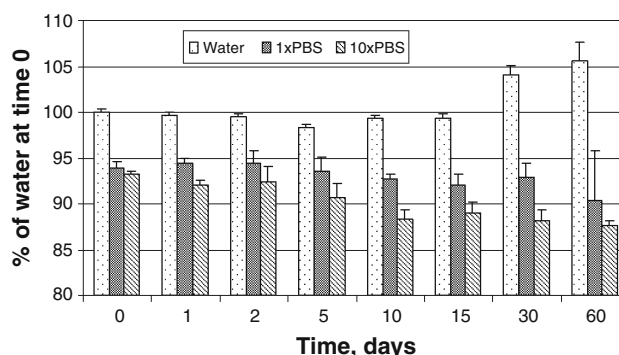
10 mL of water with soil used as a blank. Extraction and analysis of the aqueous phase using the HPLC method was performed after 1, 4 and 8 days.

Although many home care products contain PHMB, a wound care dressing was used to estimate the potential leaching of PHMB into the environment. Gauze with 0.2% PHMB, or gauze from the same manufacturer without PHMB, was cut into small pieces (approximately 1 cm). In a polypropylene test tube, 0.5 g of the gauze was placed with each of the three soil types (potting, clay or sandy) in triplicate with a no soil control. 10 mL of water was added to each sample, vortexed, and left at room temperature for a week. The aqueous portions of these samples were extracted and analyzed using the HPLC method. Because only the sandy soil contained any PHMB, the experiment was re-run with only the sandy soil and the amount of PHMB determined after 24 h.

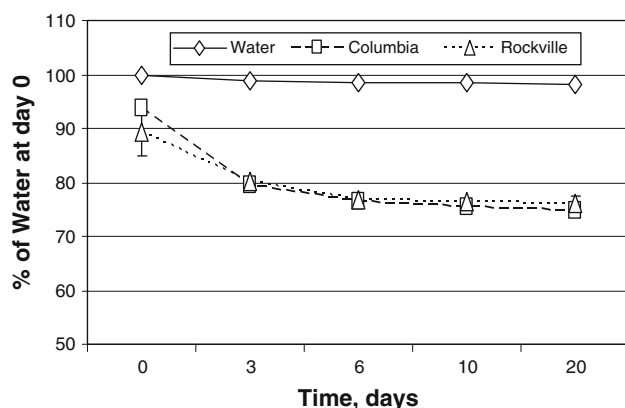
## Results and Discussion

The stability of PHMB in pure water, 1× and 10× PBS is shown in Fig. 1. There is essentially no hydrolysis of PHMB after 60 days in pure water, and 90% of the PHMB remains in both the 1× and 10× PBS. The increase in the amount of PHMB seen at the 30 and 60 day time points is due to evaporation of water; the polypropylene test tubes used were not air tight. The rate of hydrolysis in real stream water was slower, with 75% remaining after 20 days (Fig. 2). After 20 or 30 days, the amount of PHMB increased, likely due to evaporation. As expected, PHMB was completely hydrolyzed immediately when placed in HCl from 1.0 to 0.1 N, but took 30 days to degrade in 1 N NaOH (data not shown) (Kurzer and Pitchfork 1968).

The hydrolysis of PHMB does not follow a simple first-order rate because PHMB is not a single compound. PHMB is a series of polymers with a repeating hexamethylene biguanide unit  $N = 10\text{--}12$  with a range of 2–30. PHMB



**Fig. 1** The percentage of PHMB remaining in water, 1× PBS, or 10× PBS over 60 days. Data is for the 10 µg/mL level



**Fig. 2** The percentage of PHMB remaining in spikes surface water taken from local creeks. Data is for the 10 µg/mL level

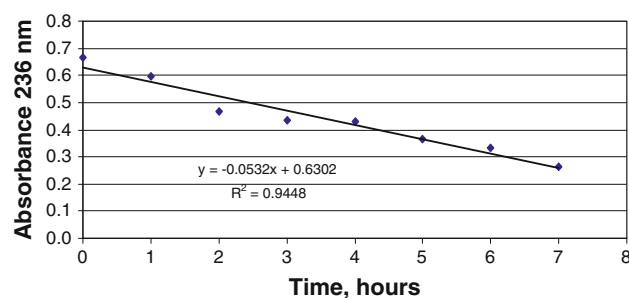
does not have a single identical end group, but could have any combination of four different end groups (Kaehn 2010).

PHMB was not detected after being placed with either the organic potting soil or the clay soil. This was expected as soil has a negative charge and binds cations tightly (Nicholls 1988). Although all the PHMB was recovered from the sandy soil after 1 day, only approximately 45% of the PHMB was available in the water fraction after 4 days and at 8 days (data not show). The surface of gravel and sand does contain minerals which may cause some degradation or binding.

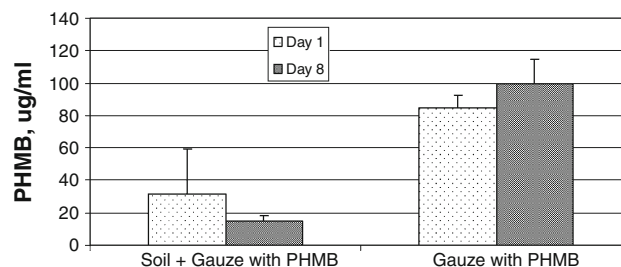
Because PHMB is so stable in water, and soil with any humic matter will strongly bind cations, the question of the stability of PHMB in water remained. As much of the water carried to streams is in concrete storm drains, the possible interaction of weathered concrete with PHMB was assessed. As seen in Fig. 3, concrete rapidly absorbed PHMB. After 24 h only about 5% (0.5 µg/mL) of the PHMB remained (data not shown). The examination of concrete as an urban sink for pesticides also showed negligible amounts of herbicides recovered from concrete run-off (Stier et al. 2005). The absorption of PHMB by concrete is not unexpected as concrete is alkaline (Lea 1998). Weathered concrete was selected to test rather than fresh concrete as the properties of concrete change over time (Lea 1998). Interaction of asphalt and concrete with human applied or inappropriately disposed chemicals is an area for further study.

The addition of PHMB impregnated gauze was readily extracted into water. Even with the addition of sandy soil, PHMB was still present in the aqueous fraction (Fig. 4). Proper disposal of PHMB containing material, such as in a land fill or in an incinerator, would probably not present any contamination to the environment.

PHMB is remarkably stable in water, however the material on which the water resides does have a significant



**Fig. 3** The absorption of PHMB into a weathered concrete sample. Data is for the 100 µg/mL level



**Fig. 4** The amount of PHMB leached from a commercially available gauze product impregnated with PHMB in water only, or in water containing 0.5 g of a sandy soil

affect on the fate of PHMB. The probability of PHMB leaching into ground water where any soil is present with any significant amount of humic matter is negligible. Microbial degradation of PHMB would suggest that this compound would serve as a nitrogen source for bacteria (O'Malley et al. 2006). The presence of PHMB in medical devices, such as wound dressing, does not present a significant threat when in a standard landfill. This may present problems to an aquatic environment if pools are drained or materials containing PHMB are placed directly into sandy streams or through storm drains. However, as concrete is a sink for this biocide, it is not likely that a significant amount of PHMB would reach a stream, unless water drained directly into the stream. The major are of concern would be the discharge of PHMB into the water of a large lake or ocean, either accidentally or purposely such as in ballast water (US Coast Guard 2004).

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