| Table 5-4. Assessment of 6PPD q sample collection methods | | | | | | | | |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Method name | Collection type | Sample type | Media | Description | Pro | Con | Example References | Comments | |
| Grab Sample | Manual | Discrete | Water | Collecting samples at a discrete time point from shore, boat, or bridge. Certified amber glass bottles are recommended. PTFE tubing and containers can be used for short-term storage (Hu et al. 2023). Some studies pre-rinse uncertified, nonglass bottles with methanol prior to collection (Rauert et al. 2022). | Accessible and low technology and cost option that offers a lot of flexibility. An amber glass bottle can be used to minimize loss of 6PPD‑q. | Transport of 6PPD‑q from surfaces occurs during storm events, some waterbodies are flashier than others making it easy to miss the 6PPD‑q peak concentration. Stormwater sampling is logistically challenging and takes a lot of boots in the field chasing storms at all hours and days of the week. The mass loading of 6PPD and 6PPD‑q is on and along roadways and stormwater infrastructure that are inherently dangerous areas to sample without proper safety precautions. Sampling near and within transportation and stormwater infrastructure often requires permission and coordination. Sampling in and around streams can also present dangerous conditions and ecological impacts (the spread of invasive species). | (Tian et al. 2021; 2022; Rauert et al. 2022; Challis et al. 2021; Lane et al. 2024; Nedrich 2022; Holzer 2023; Smith 2023) | Samples can be collected with a variety of water sampling devices depending on the target environment, including a sampling pole or bottle harness. Deeper water can be sampled using self-flushing sampling bottles on a cable triggered by a messenger. | |
| Portable Sampler | Active | Sequential | Water | Collecting samples using an autosampler from shore, many autosamplers can be programmed to take multiple samples at set intervals and duration. | The sampler can be programmed to sample throughout a storm event to capture the pollutagraph when coupled with flow measurements. Sequential sampling provides information on when 6PPD‑q is transported to waterbodies and how long it persists (resident time) by collecting samplers, for instance, every hour into separate sample containers. Portable and programmable samplers are a good tool for measuring contaminants associated with storm events to identify hotspots and determine the effectiveness of stormwater management and other actions to reduce toxicity. | Procurement, operation, and maintenance of portable samplers can be time costly. Theft and vandalism of deployed equipment in urban areas is a concern. The equipment necessary to set an automated sampling criteria adds another layer of complexity to the deployments. There is a chance that the portable sampler will malfunction, and the samples won't be collected. Sequential, discrete samples over each storm can add up to 24 samples per site, increasing the project’s analytical costs. | (Johannessen, Helm, and Metcalfe 2021; Cao et al. 2023; Tian et al. 2022) | None. | |
| Portable Sampler | Active | Composite | Water | Collecting samples using an autosampler that combines interval samples into one large container or jar. | The sampler can be programmed to sample throughout a storm event and combine the samples taken, for instance, every hour into one composited container providing an average concentration over time. Sample composites helps keep analytical costs down, especially during the screening stage of a new contaminant. | The temporal resolution of 6PPD‑q concentration trends throughout a storm event is lost. The portable samplers need to be procured, operated, and maintained by trained technical staff. The equipment needs to be secured in the field to minimize theft and vandalism. | No reference available | None. | |
| Bailer | Manual | Discrete | Groundwater | A portable grab sampler for measuring groundwater. | Relatively simple and inexpensive method for measuring groundwater once a sampling well is established. | Need to establish a sampling well. Groundwater sampling often requires technical staff and equipment. | (Zhang et al. 2023) | The modeled physicochemical properties suggest minimal movement through soils because 6PPD and 6PPD‑q have an affinity for sorbing to soils; research is needed to verify the modeled information. | |
| Grab Sample | Manual | Discrete | Sediment and soils | Collecting samples at a discrete time point from shore, boat, or bridge using a scoop. | Accessible, low-technology, and low-cost option that offers a lot of flexibility. Fewer opportunities to lose 6PPD‑q during the sampling process. | Considerable transport of 6PPD‑q from surfaces occurs during storm events. During storms the surface sediments are dynamic and may or may not represent the most recently deposited particles. | (Cao et al. 2022): Soils |  | |
| Grab Sample | Intermediate device | Discrete | Sediment | Collection of sediments using specialized devices including box corers, grabs, etc. | Allows the collection of sediments from deeper, more stable benthic environments. Grab devices or box corers come in a variety of sizes and can help standardize the sediment collection process. The sampler can homogenize large grabs and take replicate samples. 6PPD and 6PPDq have been shown to readily sorb onto particles. Collecting and measuring 6PPDq will help us further understand the contaminants’ fate and transport. | Sediment grab devices work best off boats. Larger devices require specialized winches or davets and trained technical staff. | China (Pearl River Delta, Pearl River Estuary, South China Sea): (Zeng et al. 2023)  United States and Canada: (Wu et al. 2023) | None. | |
| Core Sample | Coring device | Composite | Sediment | Collection of a sediment core that can be sectioned and possibly dated using geochronology. | Stable benthic environments can provide a deposition history from pre-industry to post-industry conditions. Sediment cores might be a good tool. Most suitable in lakes or backwater areas. | Coring devices often require technical watercrafts and personnel. The collection and processing of cores is time intensive. Coring works best in stable depositional environments such as lakes and not as well in hydrologically dynamic environments such as rivers and streams or tidally active environments. | (Nipen et al. 2022) - not specific for 6PPD‑q, it is a general reference for sediment coring and emerging organic contaminants | None. | |
| Mobile Centrifuge | Active sampler | Composite | Sediment | Collection and consolidation of particulates from water. | 6PPD‑q has been shown to readily bind to soils and sediments; therefore, high-volume collections of ambient water and consolidation could be an effective method for measuring 6PPD‑q where it exists in trace amounts. | Mobile centrifuging requires specialized equipment and trained technical field and lab staff. You can only measure one site at a time per mobile centrifuge, making site comparisons under similar conditions challenging and time consuming. | No reference available | High volume centrifuging may be a useful tool for screening sites for 6PPD‑q between and during storms. More research is needed. | |
| Sediment Trap | Sediment sampler | Composite | Sediment | A sediment trap collects modern sediments that are suspended or resuspended in the water column and deposited to the benthic environment. | Sediment traps can be deployed for days to weeks to capture the particulate sedimentation that is washed into waterbodies during storms and eventually settles. 6PPD‑q has been shown to readily sorb to particulates. Deploying, capturing, and measuring these settled particles may provide a tool for understanding the mass loading. | High-carbon environments make sediment traps challenging to deploy for longer durations because of bacterial activity and the breakdown of carbon that may release the sorbed 6PPD‑q. | No reference available | Sediment traps show promise for being a useful tool for estimating particle-bound 6PPD‑q. More research is needed to verify this method. | |
| SPME, PE, and POM | Passive sampler | Composite | Water or sediments | SPME samplers are made of fiber-optic cable with a glass core surrounded by PDMS that is absorptive to many hydrophobic chemicals. PE and POM samplers are made of organic polymer, or more simply a piece of plastic. | The analytical costs of passive samplers are often lower than active and discrete sample analysis. The samplers can be deployed for days to weeks and provide a greater chance of measuring contaminants over several storm events. The sampler targets the dissolved form of a contaminant that is the most bioavailable. 6PPDq is hydrophobic and may adhere to this media. If a model was developed, this method could be used to compare field concentrations with risk-based values or criteria. Passive samplers can be deployed in freshwater or saltwater environments. Sampling methods for 6PPDq in marine matrices is continue to develop. | Passive samplers provide an average concentration over time. They are most helpful when the rates of absorption have been estimated and a model verified to provide an estimate of concentration. A model for 6PPD‑q has not been established to date. These types of passive samplers uptake hydrophobic contaminants until an equilibrium is reached. The small amount of data available for 6PPD‑q through a storm event supports spikes in concentrations correlated with storm events. SPME passive samplers do not represent the maximum concentration that coho salmon and other sensitive species are exposed to in the environment, but rather a time-weighted average of the bioavailable 6PPD‑q. These materials can be difficult to work with in the field and lab. | (Chow et al. 2019)  For equilibrium process: (Schwarzenbach, Gschwend, and Imboden 2003)  For passive sampler methods: (Hawthorne et al. 2005; Hawthorne, Miller, and Grabanski 2009; Burgess et al. 2011; Mayer et al. 2000; Vinturella et al. 2004) | The data processing often takes technical staff to process. Some lab contractors provide data analysis and modeling services, but this increases the cost per sample. | |
| SPMD | Passive sampler | Composite | Water | A PE passive sampler that contains a lipid, used to measure trace levels of organic compounds. | Provides a time-weighted average for nonpolar or hydrophobic organic contaminants with Kow> 3. Avoids logistically challenging storm chasing. Works best in high-flow environments. | The time-weighted average may miss the 6PPD‑q concentration peaks that are helpful to understand the exposure risk and impact to sensitive species such as coho salmon. There is no absorption model for 6PPD or 6PPD‑q. Passive sampler deployments often require boats and technical field staff and modeling data analysis. | General method: (Huckins, Tubergen, and Manuweera 1990; Huckins, Petty, and Booji 2006; Schubauer-Berigan, Foote, and Magar 2012)  No references to date for studies specific to 6PPD‑q.  Research is needed to compare passive sampler effectiveness | SPMDs to measure and compare 6PPD‑q before and after toxic reduction actions or to compare sites within the same region (with similar environmental conditions) are worth investigating. | |
| POCIS | Passive sampler | Composite | Water | Microporous (0.1 µm pore size) polyethersulfone membrane encasing a solid-phase sorbent (Oasis HLB) that retains sampled chemicals. The Oasis HLB is a universal solid-phase extraction sorbent widely used for sampling a large range of hydrophilic to lipophilic organic chemicals from water. | Passive samplers help avoid storm event sampling that can be logistically challenging. The analytical costs are often lower than more traditional discrete sampling. It can provide time-integrated data to compare sites and treatment types. | Sampling rates have not been determined for 6PPD‑q. The deployment, retrieval, and data interpretation require technical field and lab staff. | (Johannessen and Metcalfe 2022) | WA department of ecology is currently evaluating the effectiveness of POCIS to detect 6PPD‑q across sites and toxic reduction actions. | |
| DGT Device | Passive sampler | Composite | Water | A passive sampler that is widely used to measure contaminants in freshwater and marine water and to assess soil and sediment. It is a low-cost plastic device that mimics biological uptake. | These samplers can be deployed for days to weeks at a time, avoiding storm-event sampling. They provide a time-weighted average concentration for select contaminants. DGT is considered a dynamic rather than an equilibrium method because it continuously removes analyte from the sampling media. They can be used in water and sediments. | This device has not been tested for measuring 6PPD‑q to date. A sampling rate for 6PPD‑q has not been estimated. | No reference available | DGT devices are often used to probe for chemical and biological processes. There is no diffusive coefficient available for 6PPD or 6PPD‑q. It uses Fick's law of diffusion. | |
| Remote Samplers, In situ | Active sampler | Continuous | Water | These devices actively pump water through an SPE media, in a stainless or PE casing, in situ for several hours per filter. Another type fills a carboy container. | These samplers can be deployed remotely and left for several hours. The filter version has no water sample to deal with, and the filters can be frozen until ready for extraction and analysis. These devices can be secured on a remote mooring. | These samples tend to get clogged after several hours and may not last for an entire storm event. The device could malfunction and miss the sampling window. | No reference available | WA department of ecology is currently evaluating the effectiveness of remote in situ active samplers to detect 6PPD‑q across sites and toxic reduction actions. | |
| B-IBI | Manual collections | Bioassessment | Biota | Benthic invertebrates are collected and identified. If the taxonomy reveals only tolerant species composition and the absence of sensitive species, then there is some sort of water quality impact. | B-IBI is the standard for measuring stream health and might be useful to correlate with 6PPD‑q as an indicator of stormwater impacts. | B-IBI is difficult to pinpoint as one disturbance among many. | (Larson et al. 2019) | WA Department of Ecology and a network of partners regularly use B-IBI to estimate the overall health of a water body. | |
| *In vitro* Using Primary Cells | Manual, active or passive collections | Bioassay | Biota | Cells isolated from an organism that are subsequently cultured and aliquots frozen (−80°C or liquid nitrogen vapor phase) for longer-term sample testing. | • Each cell line is representative of one individual. • Cell lines contain normal genetic structure. • Genetic diversity and biological replication can be increased by obtaining cells from more than one individual. • Contains multiple cells types more representative of in vivo compared to immortalized cells. | • Requires sacrificing or invasively sampling more than one animal to obtain cells over time. • Requires access to living animals. • Ambient samples can harm cell viability and must be treated prior to use in an assay (ISO protocol). • More time intensive and less consistent than immortalized cells. | Fibroblasts from killer whale: (Yajing et al. 2018)  Liver cells from mice: (Arora et al. 2009) | [ISO 5667-16:2017](https://www.iso.org/obp/ui/#iso:std:iso:5667:-16:ed-2:v1:en) provides guidance on standardizing biological tests for evaluating the effect of chemical substances on test organisms. | |
| *In vitro* Using an Immortalized Cell Line | Multiple options | Bioassay | Biota | Primary cells that do not undergo senescence and can be propagated indefinitely. Immortalized cell lines are available for many species and tissue types. | • Simplified model compared to in vivo. • Greatly reduces use of animals in testing. • Simple for many biological laboratories to integrate. • Easy access through repositories such as ATCC. • Well-established assays available to assess cell health. | • Aberrant genetic structure. • No biological replication. • Ambient samples can harm cell viability and must be treated prior to use in an assay (ISO protocol). • May not retain all physiological functions of the original cell. • Not available for all species/tissues. | RTgill-W1: (OECD 2021)  Embryonic cells isolated from coho salmon: (Greer et al. 2023) | None. | |
| *In vivo*, In Situ | Multiple options | Bioassay | Biota | Direct testing of live animals in the area under investigation. | • Direct field assessment of the environmental conditions experienced by organism. | • Many uncontrolled variables (pH, temperature, etc.) compared to laboratory studies. • Logistically challenging. | Caged fathead minnow: (Ankley et al. 2021) | None. | |
| *In vivo*, Laboratory | Multiple options | Bioassay | Biota | Testing of animals in the laboratory using field-collected water samples. | • Represents the whole conduit of chemicals present in stormwater. • Interactions between organ systems are maintained. | • Requires access to living animal. | Methods like [WET testing](https://www.epa.gov/cwa-methods/whole-effluent-toxicity-methods) in rainbow trout and other species (40 CFR 136.3) | None. | |
| *Ex vivo*, Laboratory | Multiple options | Bioassay | Biota | Extraction and isolation of tissues and cells from field-exposed animals for subsequent laboratory exposures. | Allows for the evaluation of effects in wild species exposed to a stimulant introduced in a controlled manner. | Intricate procedures require highly trained personnel. | Short-term culture and stimulation of immune cells (Rehberger et al. 2021) | None. | |
| Biota Sampling | Collection and processing of organism tissues and plasmas (e.g., fish tissues, blood and bile, mussels, plants, invertebrates, microbes) | Composite | Biota | The collection and analysis of biota tissues provides indicators of the magnitude of toxic contamination within the species’ geographic habitat and help us understand the bioaccumulation dynamics. | Fish and mollusks can provide a direct measure of 6PPD and 6PPD‑q bioavailability in surface water bodies. For example, 6PPD and 6PPD‑q have been detected in snakehead, weever, Spanish mackerel, English sole, and mussels. | Fishing often requires specialized electro fishing equipment and technical field and lab staff to collect and process samples. | Ji et al. 2023 | Fish and mollusk tissue provide a promising method for detecting bioavailability of 6PPD and 6PPD‑q in aquatic environments, but more research is needed. | |
| Ambient Air Sample | Active | Composite | Air | Large-volume air sample collected on a quartz GFF over a relatively long duration (~24-hours). Cascade impactor can be used in sample collection, which would allow for determining concentrations in different size fractions (e.g., PM2.5). | Captures particle-bound fraction. If a cascade impactor is used, the concentration in different size fractions can be determined. | Due to relatively low concentrations in air, large sample volumes are required. Monitoring is necessary to ensure sample integrity and security. Sampling equipment can be large and noisy because of the need for a high flowrate fan. A continuous power supply is needed. Would not capture all of the fraction that may be present in the vapor phase. | Cao et al. 2022, Zhang et al. 2022a, Zhang et al. 2022b | Researchers have reported 6PPD and 6PPD‑q have low vapor pressures and are not likely to volatilize at 25°C, but significant data gaps exist concerning their presence in the vapor phase. | |
| Ambient Air Sample | Passive | Composite | Air | Passive PUF disk samplers deployed over an extended period (e.g., months). | Samples represent the whole air mixture (i.e., particle-bound and vapor fractions). No fan is required. Lower cost and complexity compared to active air sampling. | Samplers must be deployed for extended periods. Monitoring is necessary to ensure sample integrity and security. | Gaga 2019 (general PUF reference), Johannessen et al. 2022 | None. | |
| Dust Sample | Manual | Grab | Dust | Collection of dust from roadways, parking garages, tunnels, waste management facilities, and indoor environments using a variety of collection methods, such as vacuum cleaner, brush and shovel, pressure washing, and wet vacuum. | Sample collection is relatively easy. Pressure washing and wet vacuum are reported to be more efficient in collecting smaller particles. | Dry vacuum sampling may be less efficient in collecting smaller particles. | Liang et al. 2022,  Liu et al. 2019,  Deng et al. 2022,  Hiki and Yamamoto 2022,  Klöckner et al. 2021,  Zhang et al. 2022b,  Huang et al. 2021 | There are gaps in our understanding about how the use of dry vs. wet sample collection methods could impact the transformation of 6PPD or 6PPD‑q. | |
| Grab Sample | Manual | Discrete | Snow | Collection of snow along roadways and associated particulates. | Snow captures and consolidates the TWPs and 6PPD‑q over time. Snow is relatively straightforward to collect. | Snow samples can be collected only opportunistically, and collection works better in areas with regular snow events. Method would not distinguish between contribution from road dust vs. atmospheric deposition. See comments. | (Challis et al. 2021; Maurer et al. 2023; Seiwert et al. 2022) | Studies to date have not evaluated the relative contribution in snow from potential atmospheric sources vs. road dust sources. | |

Notes: µm=micrometer, ATCC=, B-IBI=Benthic Index of Biotic Integrity CFR=Code of Federal Regulations DGT=, diffusive gradients in thin-films, GFF=glass fiber filter, PDMS=polydimethylsiloxane, PE=polyethylene, POCIS=polar organic chemical integrative sampler, POM=polyoxymethylene, PTFE=polytetrafluoroethylene, PUF=polyurethane foam, SPMD=semi-permeable membrane device, SPME=solid-phase micro-extraction, TWP=tire-wear particles, WET=whole effluent toxicity

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