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RADIOCARBON DATING AND FRESHWATER RESERVOIR EFFECTS OF AQUATIC MOLLUSKS WITHIN FLUVIAL CHANNEL DEPOSITS IN THE MIDWESTERN UNITED STATES

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ABSTRACT. Late Quaternary fluvial channel deposits are notoriously difficult to date. In the midwestern United States, shells of aquatic mollusks can be found within many fluvial channel sediments and therefore can be radiocarbon (^{14}C) dated to determine the age of the deposits. However, carbonate platform rocks are abundant in this region, potentially causing freshwater ^{14}C reservoir effects (FRE) in mollusk shells. We ^{14}C dated 11 aquatic gastropod and bivalve shell samples from specimens collected live from a stream in southwestern Ohio during three different years to assess the modern ^{14}C reservoir effect. Modern samples yielded an average ^{14}C FRE_{modern} of 518 ± 65 ^{14}C yrs for 2020 ($n=5$), 640 ± 34 ^{14}C yrs for 2021 ($n=2$), and 707 ± 76 ^{14}C yrs for 2022 ($n=4$). We also ^{14}C dated matched pairs of organic wood or charcoal and aquatic mollusk shells from late Pleistocene and Holocene deposits in the Four Mile Creek floodplain to determine the FRE_{fossil}. These samples, free of any potential influence from nuclear bomb testing, yielded an overall weighted mean FRE_{fossil} of 1029 ± 345 ^{14}C yrs. We then assess the advantages and limitations of both the FRE_{modern} and FRE_{fossil} methods for determining freshwater reservoir effects. Finally, we apply the FRE_{fossil} correction to a series of shell ages from fluvial terrace deposits as a case study. The results indicate that although there is a ^{14}C FRE in streams from the midwestern United States, aquatic shells can provide robust age control on fluvial channel deposits. More research is needed to understand the spatial and temporal variability of FREs, as well as any species effects, among various watersheds across the midwestern United States.

KEYWORDS: bivalve, freshwater reservoir effect, fluvial deposits, gastropod, geochronology, mollusks, radiocarbon, rivers, streams.

INTRODUCTION

Stream valleys are one of the few places on landscapes where there is typically a long history of sedimentation, and consequently, provide the potential to record extensive sequences of environmental change. As such, geomorphologists use fluvial deposits to identify climatic (Waters and Haynes 2001; Tully et al. 2019), tectonic (Burnett and Schumm 1983; Burbank et al. 2012), and anthropogenic (Knox 2001, 2006) drivers of regional landscape evolution. In the midwestern United States, glacial outwash systems excavated deep (~50–150 m) river valleys during the Pleistocene that later filled with thick sequences of fluvial deposits. These deposits, however, have not been used widely for reconstructing late Pleistocene and Holocene environmental change due, in part, to the difficulty in dating these deposits.

Geomorphologists have traditionally focused on radiocarbon (^{14}C) dating fluvial overbank deposits, which sometimes contain wood, charcoal, and buried soil organic horizons, to constrain the age of fluvial sequences (e.g., Waters and Haynes 2001; Tully et al. 2019). Fluvial terraces, however, can still receive episodic overbank sedimentation long after the floodplains were initially abandoned, thereby only providing a minimum age of the underlying channel sediments. Moreover, fluvial channel sediments are often critical for understanding changes in the nature of stream systems over time. Many late Pleistocene and Holocene channel deposits in the midwestern United States contain shell fragments of aquatic mollusks (Figure 1). These shells can be ^{14}C dated, but the dissolution of widespread carbonate rocks may cause ^{14}C freshwater reservoir effects (FRE) due to the addition of ^{14}C -depleted inorganic carbon to streams.

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Figure 1 Late Holocene fluvial channel deposits, approximately 4000 years old, with aquatic mollusk and terrestrial gastropod shells from the Four Mile Creek floodplain, Ohio.

Radiocarbon reservoir effects in aquatic organisms, which cause ^{14}C ages to be too old, occur when the dissolved inorganic carbon pool of the water body is not in equilibrium with the atmosphere. Reservoir effects have been fairly well characterized for the marine realm (Hadden et al. 2023; Heaton et al. 2020), where ^{14}C ages are on average ~ 500 ^{14}C years too old, but can vary widely between ~ 30 to 1000 ^{14}C yrs. Research on freshwater reservoir effects (FREs) has mostly focused on lakes and paleolakes, as these represent key paleoclimatic archives (Geyh et al. 1997; Yu et al. 2007; Ascough et al. 2010; Soulet et al. 2011; Jull et al. 2014; Zhou et al. 2022). Much less research has been conducted on the magnitude and variability of freshwater reservoir effects in streams (Philippsen 2013; Philippsen and Heinemeier 2013; Osterkamp et al. 2014; Coularis et al. 2016), especially on mollusk shells. As a result, the potential for using aquatic mollusk shells for the ^{14}C dating of fluvial deposits has been limited.

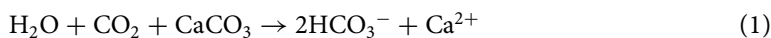
Here we use two different methods to assess the ^{14}C FRE of Four Mile Creek, a small watershed with carbonate bedrock located in southwestern Ohio, USA. First, to determine the FRE of the modern stream system, we measured the ^{14}C activity of mollusks collected live from the stream in 2020, 2021, and 2022. Second, to determine the FRE of the late Pleistocene and Holocene stream system, we ^{14}C dated matched pairs of wood or charcoal organic materials and aquatic mollusk shells from fluvial terrace deposits. We then calculate the FRE and propagate the error for each method, and discuss the advantages and limitations of each method for determining the FRE. Finally, as a case study, we apply the FRE for Four Mile Creek to determine the age of a series of fluvial terrace deposits to demonstrate the potential and limitations of using aquatic mollusk shells to provide age control for fluvial deposits or archaeological shell middens (e.g., Peacock et al. 2005; Genheimer and Hedeem 2014).

Freshwater Reservoir Effects in Streams

Freshwater reservoir effects occur when there is an input of ^{14}C -depleted carbon to the watershed that is not in equilibrium with the atmosphere. There are several potential sources of old carbon, including both organic and inorganic. Organic carbon sources include the decomposition of buried organic material from peats, paleosols, and floodplain deposits, as

well as organic carbon from bedrock such as coals or black shales. In most watersheds, the decomposition of organic carbon is unlikely to cause significant reservoir effects due to the low abundance of organic carbon. Significant reservoir effects, however, can arise due to inorganic carbon sources such as volcanic or geothermal degassing into shallow groundwater aquifers or lakes, as well as due to carbonate bedrock such as limestone (CaCO_3) or dolomite ($\text{CaMg}(\text{CO}_3)_2$). Reservoir effects are especially pronounced when there is limited vegetation on the landscape to pump atmospheric CO_2 into soils or waterbodies. For example, stream water from the Rio Loa in the hyperarid Atacama Desert of northern Chile was analyzed for dissolved inorganic carbon (DIC) and was found to have <27 percent modern carbon (pMC), equating to a >11,000 ^{14}C yr offset from modern (Aravena and Suzuki 1990). The discharging of fossil aquifers, or long travel times for groundwater flowpaths to streams, will also exacerbate FREs due to the decay of ^{14}C subsequent to groundwater recharge (e.g., Riggs 1984; Fontes and Gasse 1991; Osterkamp et al. 2014).

In streams, especially in humid environments, freshwater reservoir effects are more often due to the dissolution of carbonate rocks, which cover ~15% of Earth's terrestrial landscape (Goldscheider et al. 2020). Even when carbonate rocks occupy only a small percentage of the landscape, the high dissolution rate of carbonate minerals allows these rocks to control the aqueous chemistry of the watershed. The dissolution of limestone occurs as CO_2 dissolves in water creating carbonic acid which then dissolves carbonate rock (1):



In most watersheds, carbonic acid is present in rainwater and is also produced in soils as plants pump CO_2 into the vadose zone during the growing season, increasing soil CO_2 concentrations over atmospheric concentrations by an order of magnitude (Quade and Cerling 2006). As carbonic acid is required for carbonate dissolution, and carbonic acid is produced from atmospheric CO_2 in most settings, the highest concentration of DIC originating from limestone in stream water is 50% (Broecker and Walton 1959; Philippsen 2013). Therefore, the maximum FRE is ~5730 ^{14}C yrs, or one ^{14}C half-life, in watersheds with carbonate rocks unless there is an alternate source of ^{14}C -depleted CO_2 .

Research on freshwater reservoir effects began with lakes, and little work has been conducted on freshwater streams. The presence of reservoir effects in lakes within limestone terrains was first suggested by Godwin (1951), and then demonstrated in lakes in the northeastern United States by Deevey et al. (1954) and in streams and lakes in the Great Basin, USA (Broecker and Walton 1959). Deevey et al. (1954) and Broecker and Walton (1959) identified reservoir effects of up to several thousand years on DIC in lakes, whereas DIC from streams draining silicate rocks were found to be in equilibrium with the atmosphere. Two subsequent studies on FREs in streams have examined the ^{14}C content of DIC. Philippsen and Heinemeier (2013) determined the ^{14}C activity of DIC, among other materials, on the Trave and Alster Rivers in northern Germany, with watersheds dominated by glacial till that contain ~20% carbonate minerals. The FRE for the Alster River ranged from 1873 ± 47 to 3044 ± 57 ^{14}C yrs ($n=4$) and the Trave River ranged from 1528 ± 45 to 2634 ± 41 ^{14}C yrs ($n=4$). These results indicate fairly high FRE values, especially for a watershed in a humid setting and still influenced by post-bomb ^{14}C . Coularis et al. (2016) analyzed the ^{14}C activity of 49 DIC samples from 20 sampling sites on the Loire River in France. In the majority of the study region FRE values were mainly between 135 and 667 ^{14}C yrs. In the region with the most limestone, however, FRE values ranged from 690

to 2251 ¹⁴C yrs. The authors found a good correlation between FRE values and stream alkalinity (Coularis et al. 2016).

The FRE determined from the ¹⁴C activity of DIC, representing one snapshot in time, is fundamentally different than the FRE of aquatic mollusk shells, which represent a time-averaged FRE during the growth of the shell. Two mollusk shells analyzed from the Alster River by Philippsen and Heinemeier (2013) yielded FRE values of 869 ± 34 and 1654 ± 35 ¹⁴C yrs, both significantly lower than FRE values derived from DIC. Osterkamp et al. (2014) analyzed ~30 matched pairs of ¹⁴C ages on aquatic bivalves and charcoal from late Pleistocene and Holocene archaeological sites along the Snake River in northwestern United States and found FRE values of ~2500–3000 ¹⁴C yrs for most samples. The authors attributed the reservoir effects in this semi-arid environment with a basaltic aquifer system to long travel times for groundwater discharging as base flow in the Snake River (Osterkamp et al. 2014). Active geothermal activity may also play a role in this region.

The majority of research on ¹⁴C reservoir effects in streams has come from Europe and Asia with work focused on the impact of FREs on fish or other animals that were consumed by humans (Cook et al. 2001; Philippsen 2013; Philippsen and Heinemeier 2013; Piličiauskas and Heron 2015; Kuzmin et al. 2020; Schulting et al. 2022). In general, this is an extremely difficult question as large freshwater fish can live for decades and move from streams to lakes as well as to estuaries, depending on the watershed. Aquatic mollusks have a much simpler, and sessile, life history making their shells a more suitable material for ¹⁴C dating.

STUDY AREA

Four Mile Creek is an ~432 km² watershed situated in the states of Ohio and Indiana in the midwestern United States (Figure 2). This region receives ~105 cm/yr precipitation and is characterized by a temperate deciduous forest, although most of this region has been deforested for agriculture. Precipitation is well distributed throughout the year with all months receiving >70 mm, but the spring and early summer are the wettest seasons averaging ~100 mm/mo (Figure 3). Mean annual stream discharge has been estimated to be ~5.5 m³/s where Four Mile Creek exits its watershed and combines with the Seven Mile Creek watershed (Rech et al. 2018). Although precipitation is well distributed throughout the year, there is a pronounced dry season in Four Mile Creek during the latter half of summer and early autumn due to both diminished rainfall and high evapotranspiration rates (Figure 3).

Bedrock in the watershed is comprised of Upper Ordovician shale with ~25–50% interbedded limestone beds that are typically 5–25 cm thick (Swinford and Schumacher 1997; Swinford and Vorbau 1999). In the upland regions the Ordovician shale is covered by ~2–5 m of fine-grained glacial till that dates to ca. 24 ka during the Last Glacial Maximum (Ekberg et al. 1993). The valley fill in Four Mile Creek is ~50 m thick and contains interbedded sequences of glacial till, glacial outwash, proglacial lacustrine deposits, and fluvial sediments. The glacial till is composed of ~15–20% carbonate minerals and clasts of limestone are common within the glacial outwash. Modern-day Four Mile Creek is primarily a bedrock stream in the upper reaches with thin and discontinuous deposits of alluvium above the Acton Lake reservoir. Below Acton Lake, Four Mile Creek is predominately an alluvial stream with a broad floodplain (~1 km wide) with only intermittent exposures of glacial till and underlying Ordovician bedrock. There is one reach of Four Mile Creek near Lane's Mill, however, where the stream is deeply incised into Ordovician bedrock (Tenison 2022) (Figure 2).

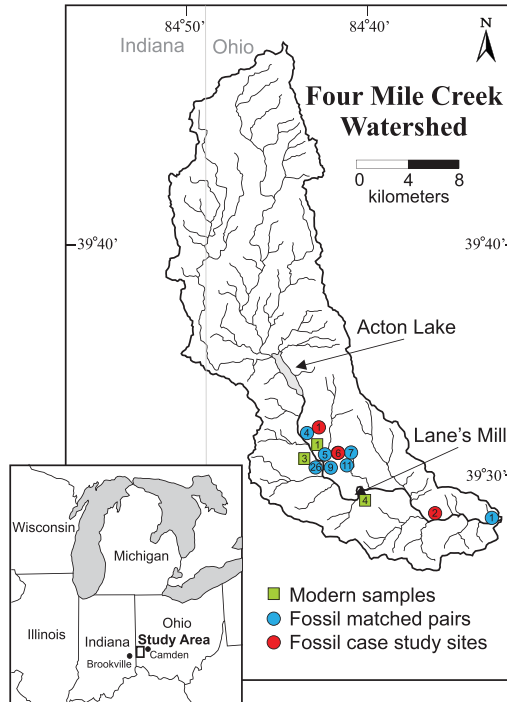


Figure 2 The Four Mile Creek watershed in southwestern Ohio and southeastern Indiana, USA, with the approximate location of modern and fossil samples.

Today Four Mile Creek is a meandering stream system characterized by riffle-pool sequences and large gravel bars of limestone cobbles and flagstones (Rech et al. 2018). The Four Mile Creek floodplain contains sequences of fluvial terraces that are in general between ~2 and 11 m above modern stream level (Tenison 2022). During the late summer and early autumn stream discharge drops, leaving many run and riffle sections to go dry with mostly stagnant water in pools <2 m deep. Total dissolved solids (TDS) in streams increase during late summer and early autumn due to base flow fed by groundwater containing high calcium and bicarbonate as a result of the groundwater interacting with limestone bedrock and fine-grained carbonate material within the glacial till (Figure 3).

METHODS

Collection

Modern aquatic mollusks were collected live in October of 2020 (n=5), 2021 (n=2), and 2022 (n=4) near the assumed end of the shell growth period for the mollusks. The mollusks were collected from three different sites within Four Mile Creek study area. Sites 1 and 3 are situated within pool-riffle complexes, whereas Site 4 is situated along a bedrock (shale-limestone) reach of the stream (Figure 2; Table S1). Aquatic gastropods (Physidae, possibly *Physa acuta*) and invasive aquatic clams (*Corbicula fluminea*) were collected as these were the only taxa of live specimens readily found (Table 1). The aquatic clam specimens were all collected from riffles, whereas the gastropods were collected from slower moving waters (channel margins or pools).

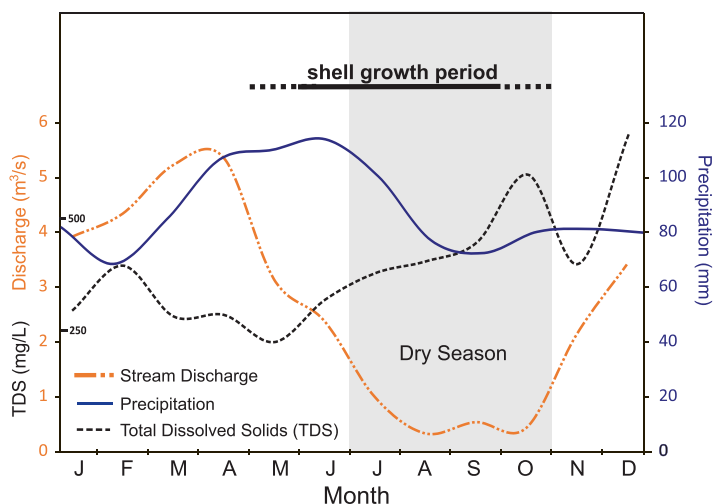


Figure 3 Average stream discharge, total dissolved solids, and precipitation for the study area. Precipitation is a 30-year average (1991–2021) from the weather station at Brookville, Indiana, situated 25 km to the southwest of the study area. Stream discharge data is a 10-year average from the USGS gaging station on Seven Mile Creek in Camden, Ohio, that is approximately 15 km northeast of the study area. Seven Mile Creek is the comparable watershed that is adjacent to Four Mile Creek and has a USGS gaging station (USGS#03272700).

Fossil aquatic mollusk shells and organic material (small pieces of wood or charcoal) were collected as matched pairs from excavated backhoe trenches or river outcrops of paleochannel deposits within the Four Mile Creek valley (Tables 2 and S1). The paleochannel deposits at these sites were all coarse-grained (pebble-sized) and likely riffle-run sequences. The fossil mollusk shell and organic material comprising a matched pair were collected from similar depths within the stratigraphic section. A larger variety of species were identified in the fossil mollusk samples compared to the modern samples, so species sampled were more diverse in the fossil record. In addition to the fossil matched pairs, three additional paleochannel deposits with fossil shells that had been ¹⁴C dated were utilized to demonstrate the impact of the FRE age correction and error on samples (Tables 3 and S1).

Chemical Pretreatment

Both the modern and fossil carbonate shell samples were soaked in 18.2 MΩ water, sonicated briefly (~3 s to 6 min total) depending on the fragility of the shell, and rinsed to remove any detrital sediment. Samples were then soaked in 10–15 mL of 5.65–6% sodium hypochlorite (NaOCl) at 60°C for >4 hr, rinsed 3 times with 18.2 MΩ water, then dried in a vacuum drying oven at 60°C overnight. Fossil carbonate shell samples were leached with 0.1 N HCl to remove the outer shell material (>20% of shell mass) that can sometimes contain secondary carbonate coatings. Samples were then rinsed 3 times with 18.2 MΩ water and dried in a vacuum drying oven at 60°C overnight. Once modern and fossil carbonate shells were dried, 15–30 mg of shell was selected and sent to Direct AMS for analysis. Modern samples were not acid leached, and no samples were powdered, to reduce the possibility of atmospheric CO₂ interacting with the increased surface area of shell material.

Modern samples of Physidae were ~5–12 mm in length. The age of the Physidae specimens at the time of collection is unknown and the entire specimen was used for ¹⁴C analysis. The

Table 1 Freshwater reservoir effects (FREs) of aquatic mollusks collected live.

Sample ID	Study site	Lab #	Taxa [^]	Collection	pMC	1 σ	Apparent ¹⁴ C age	FRE _{modern} *	1 σ
1a	1	D-AMS 041000	<i>Corbicula fluminea</i>	2020	94.61	0.23	445 ± 20	456	25
1b	1	D-AMS 040071	Physidae	2020	93.71	0.25	522 ± 21	532	27
3a	3	D-AMS 041001	<i>Corbicula fluminea</i>	2020	94.64	0.25	443 ± 21	453	27
4a	4	D-AMS 040072	<i>Corbicula fluminea</i>	2020	92.86	0.23	595 ± 20	605	26
4b	4	D-AMS 040073	Physidae	2020	93.58	0.26	533 ± 22	543	27
3_2	3	D-AMS 044981	<i>Corbicula fluminea</i>	2021	92.33	0.23	641 ± 20	616	26
4_2	4	D-AMS 044982	<i>Corbicula fluminea</i>	2021	91.78	0.22	689 ± 19	664	25
FMC St. 1 2022 1	1	D-AMS 051434	<i>Corbicula fluminea</i>	2022	91.71	0.28	695 ± 25	670	29
FMC St. 1 2022 2	1	D-AMS 051435	<i>Corbicula fluminea</i>	2022	91.42	0.28	721 ± 25	696	29
FMC St. 4 2022 1	4	D-AMS 051436	<i>Corbicula fluminea</i>	2022	91.98	0.28	672 ± 24	647	29
FMC St. 4 2022 2	4	D-AMS 051437	<i>Corbicula fluminea</i>	2022	90.05	0.30	842 ± 27	817	31

[^]Physidae: *Physa gyrina* or *Physa acuta*.

*FRE = Freshwater Reservoir Effect. The F¹⁴C (May–October) was predicted to be 1.0013 ± 0.002 in 2020, 0.9969 ± 0.002 in 2021, and 0.9926 ± 0.002 in 2022 (Hua et al. 2022).

modern *Corbicula fluminea* samples were approximately 10–20 mm in width and are thought to be <2 years old based on their size. From these modern *Corbicula fluminea* shells, small fragments (2–4 mm) from recent shell growth from multiple specimens at each locality were combined for ¹⁴C analysis.

Organic samples were manually separated from any soil material through careful selection in the lab. Larger organic pieces were then crushed or broken into smaller pieces to allow better penetration during chemical pretreatment. Organic samples then underwent standard acid, base, acid (ABA) chemical pretreatment with 1 N HCl at 60°C for at least 30 min, and this step was repeated if there was a strong reaction, followed by 1 N NaOH at 60°C for at least 30 min, and this step was repeated until the supernatant was clear or a light tea color, followed by 1 N HCl at 60°C for at least 30 min. Samples were then rinsed with 18.2 MΩ water and decanted until the pH was greater than 4. Finally, 5–10 mg of pretreated material was sent to Direct AMS for analysis (Tenison 2022).

Calculation of Freshwater Reservoir Effects: Modern Mollusks

The modern freshwater reservoir effect (FRE_{modern}) was calculated for each sample of aquatic mollusks collected live in 2020, 2021, and 2022 by comparing the percent Modern Carbon (pMC) of the aquatic mollusk shells relative to the atmosphere at the time of collection:

$$FRE_{\text{modern}} = 8033 * \ln\left(\frac{pMC_{(\text{atm})}}{pMC_{(\text{mol})}}\right) \tag{2}$$

where 8033 is the conventional Libby mean life of ¹⁴C; $pMC_{(\text{atm})}$ is the ¹⁴C concentration of the atmosphere at the time of collection for the aquatic mollusks; and $pMC_{(\text{mol})}$ is the ¹⁴C concentration of the aquatic mollusk shells.

The ¹⁴C concentration of the atmosphere at the time of sample collection was determined by extrapolation from the 1950–2019 atmospheric ¹⁴C concentration (Hua et al. 2022) for Zone 2 as defined by the authors. The atmospheric ¹⁴C content was extrapolated to be 1.0013 ± 0.002 , or 100.13 pMC, for 2020 (May–October); 0.9969 ± 0.002 , or 99.69 pMC, for 2021 (May–October); and 0.9926 ± 0.002 , or 99.26 pMC. FREs were determined separately for each year.

The uncertainties within 1 standard deviation for the FRE_{modern} of each sample of aquatic mollusk shells collected live were calculated using the following equation (Philippsen 2013: Eq. 3):

$$\sigma \text{ of } FRE_{\text{modern}} = 8033 * \sqrt{\left(\frac{\Delta pMC_{(\text{atm})}}{pMC_{(\text{atm})}}\right)^2 + \left(\frac{\Delta pMC_{(\text{mol})}}{pMC_{(\text{mol})}}\right)^2} \tag{3}$$

where $\Delta pMC_{(\text{atm})}$ is the error associated with the $pMC_{(\text{atm})}$; $\Delta pMC_{(\text{mol})}$ is the error associated with the $pMC_{(\text{mol})}$; and all other variables are used as previously defined.

The FRE_{modern} and 1 standard deviation of FRE_{modern} of each year were averaged to get the final ¹⁴C FRE_{modern} value for each year. A weighted mean was not used for these samples as the errors for individual samples were similar.

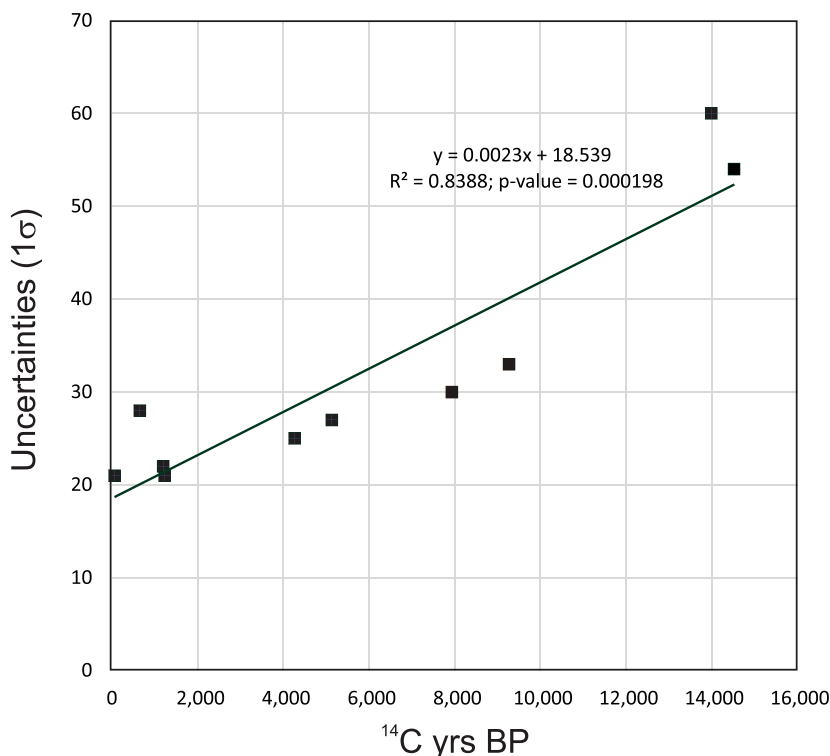


Figure 4 Plot of the uncalibrated ^{14}C ages of fossil organic and shell material versus the 1σ uncertainty of the ^{14}C age from Four Mile Creek.

Calculation of Freshwater Reservoir Effects: Fossil Mollusks

The freshwater reservoir effect for each set of matched shell-organic fossil pair ($\text{FRE}_{\text{fossil}}$) was calculated by subtracting the conventional ^{14}C age of the organic sample from the age of the shell sample. The overall fossil freshwater reservoir effect was determined by calculating the weighted mean of the fossil pair reservoir ages (e.g., Yu et al. 2018). However, before the weighted mean was calculated, age-adjustment using linear regression was used to remove the effect of the ^{14}C age on the 1σ uncertainties. This was necessary due to the large temporal span of the matched pair dataset. The weighted mean calculation weights ^{14}C ages with smaller uncertainties greater than ^{14}C ages with larger uncertainties, so it is necessary to remove this effect as otherwise there is a temporal bias in the data.

A linear regression between each ^{14}C age and its 1σ uncertainty ($x=\text{age}$, $y=1\sigma$) was calculated to determine the strength of correlation ($R^2=0.84$) and the slope ($b_1=0.0023$) for the relationship (Figure 4). The age-adjustment to remove the effect of age from the 1σ values was then calculated for each fossil ^{14}C sample using the equation:

$$\text{Age Adjusted } 1\sigma = Y - b_1 * X \quad (4)$$

where Y is the original, non-adjusted 1σ value; b_1 is the slope from the linear regression model; and X is the ^{14}C age associated with the original, non-adjusted 1σ value.

The age-adjusted uncertainty was then calculated using the propagation of uncertainty equation for subtraction:

$$\text{Age Adjusted Paired } 1\sigma = \sqrt{\sigma_A^2 + \sigma_B^2 - 2\sigma_{AB}} \tag{5}$$

where σ_A is the age-adjusted 1 σ value for the shell sample; σ_B is the age-adjusted 1 σ value for the organic sample; and σ_{AB} is the covariance variable, which was not used.

The freshwater reservoir effect (FRE_{fossil}) and age-adjusted uncertainty for each set of matched pair samples were utilized to calculate the overall combined weighted mean of the five fossil matched pair samples, i.e. the weighted mean of FRE_{fossil} (Bevington and Robinson 2003: Eq. 4.17):

$$\text{Weighted Mean of FRE}_{fossil} = \mu' = \frac{\sum_i \frac{R_i}{\sigma_i^2}}{\sum_i \frac{1}{\sigma_i^2}} \tag{6}$$

where R_i is the reservoir effect value from each matched pair; and σ_i is the age adjusted 1 σ uncertainty from each matched pair.

The uncertainty associated with the weighted mean of FRE_{fossil} was calculated by determining the weighted average of the variance of the reservoir effect (Bevington and Robinson 2003: Eq. 4.22) and square-rooting the result to get σ (standard deviation):

$$\text{Weighted } \sigma \text{ of FRE}_{fossil} = \sigma = \sqrt{\left(\frac{\sum w_i R_i^2}{\sum w_i} - \mu'^2\right) * \frac{N}{(N-1)}} \tag{7}$$

where w_i is $1/\sigma_i^2$, where σ_i is the uncertainty in each R_i value; R_i is the reservoir effect value from each matched pair; μ' is the weighted mean of FRE_{fossil} (result of Eq. 6); and N is the sample size.

RESULTS

Modern aquatic mollusk shells collected live in 2020 (n=5) had pMC values that ranged from 92.86 to 94.64 with apparent ¹⁴C ages between 443 to 595 ¹⁴C yrs (Table 1). The two Physidae shells from modern sites 1 and 4 had FRE_{modern} values of 532 ± 27 and 543 ± 27 ¹⁴C yrs, whereas the three *Corbicula fluminea* samples from sites 1, 3, and 4 had values of 456 ± 25, 453 ± 27, and 605 ± 26 ¹⁴C yrs. The average FRE_{modern} value for 2020 is 518 ± 65 ¹⁴C yrs. In 2021 two *Corbicula fluminea* shells were analyzed that had pMC concentrations of 91.78 and 92.33 and apparent ¹⁴C ages of 641 ± 20 and 689 ± 19 ¹⁴C yrs. The average FRE_{modern} value for 2021 is 640 ± 34 ¹⁴C yrs. *Corbicula fluminea* collected live in 2022 (n=4) had shell pMC values that ranged from 90.05 to 91.98 with apparent ¹⁴C ages between 672 to 842 ¹⁴C yrs (Table 1). The average FRE_{modern} value for 2022 is 707 ± 76 ¹⁴C yrs. Matched pair ¹⁴C ages of fossil aquatic mollusk shells and organics from late Pleistocene and Holocene channel deposits show that shell ages are on the order of 500 to 1300 ¹⁴C years older than organics (Table 2; Figure 5). One matched pair sample (Trench 5) displayed an organic ¹⁴C age that was ~300 years older than the shell age, which was therefore excluded from the weighted mean of FRE_{fossil} determination (Table 2). One other sample (Trench 7) had a shell ¹⁴C age that was ~3200 ¹⁴C years older than the organic age. A second shell was ¹⁴C dated from this unit, which also yielded an age ~3000 ¹⁴C years older than the organic age. Because the organic ¹⁴C age

Table 2 Freshwater reservoir effects (FREs) derived from matched pairs of fossil organics and aquatic mollusks.

Location	Lab #	Material [^]	# of shells	Depth (cm)	¹⁴ C age	1 σ	1 σ (adj) [◇]	FRE _{fossil}	FRE 1 σ [*]
Station 1	D-AMS 042277	<i>Pisidium compressum</i> , <i>Sphaerium</i> sp.	6	-235	14,533	54	21		
Station 1	D-AMS 042282	Organics		-235	13,997	60	28	536	81
Trench 11	D-AMS 042276	<i>Pisidium</i> sp.	7	-125	9274	33	12		
Trench 11	D-AMS 042281	Organics		-125	7941	30	12	1333	45
Station 26C	D-AMS 044983	<i>Sphaerium striatinum</i>	1	-105	5135	27	15		
Station 26C	D-AMS 044993	Organics		-105	4270	25	15	865	37
Trench 5	D-AMS 042274	<i>Sphaerium</i> sp.	1	-165	1496	20	na		
Trench 5	D-AMS 042279	Organics		-170	1782	20	na	-286	na
Trench 7	D-AMS 043606	Unknown bivalve	1	-150	4307	25	na		
Trench 7	D-AMS 042275	<i>Pleurocera canaliculata</i> or <i>Elimia</i> sp.	1	-150	4106	23	na		
Trench 7	D-AMS 042280	Organics		-150	1084	26	na	3022	na
Trench 4	D-AMS 042273	<i>Sphaerium</i> sp.	2	-105	1198	22	19		
Trench 4	D-AMS 036641	Organics		-105	657	28	26	541	36
Station 9	D-AMS 042278	Unidentifiable aquatic gastropods	3	-220	1233	21	18		
Station 9	D-AMS 042283	Organics		-220	62	21	21	1171	30

[^]All organic samples were charcoal or wood.

[◇]See text for explanation of adjusted 1 σ values.

*1 σ errors were only calculated for matched pairs that were used to calculate the FRE.

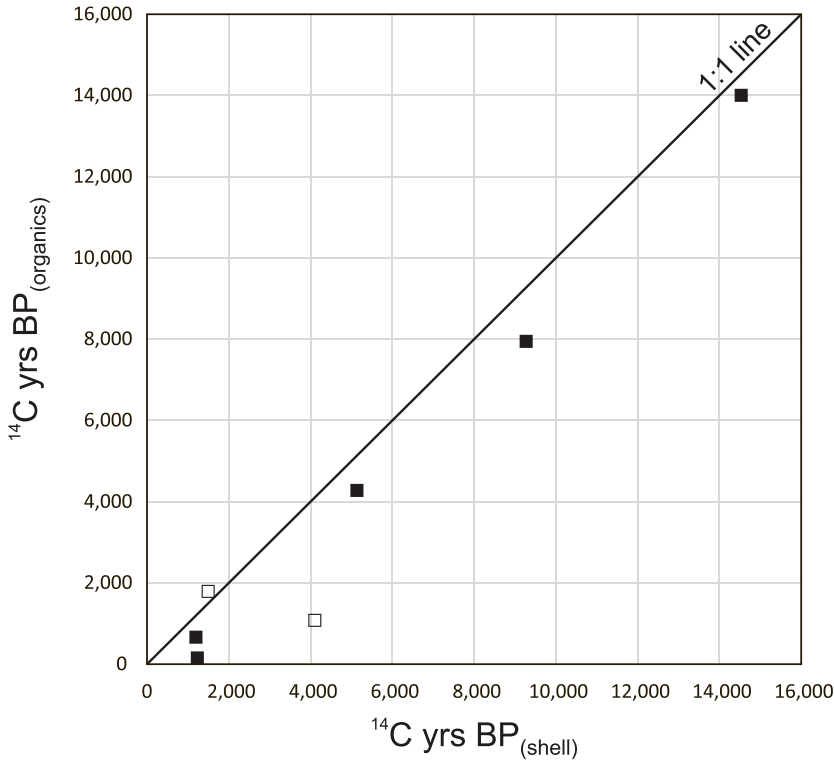


Figure 5 Plot of the uncalibrated ¹⁴C ages of matched pairs of fossil aquatic mollusk shells and organic material (wood or charcoal) from the Four Mile Creek floodplain deposits. All matched pairs plot to the right of the 1:1 line, indicating older ages of shell material, except for the matched pair from Trench 5. Open boxes signify matched pairs that were not used to determine the fossil FRE.

appeared much too young compared to the two shell ages, these matched samples were omitted from the weighted mean of FRE_{fossil} determination. The five remaining matched pairs had FRE_{fossil} values that ranged from 536 ± 81 to 1333 ± 45 ¹⁴C yrs, and have a weighted mean of FRE_{fossil} of 1029 ± 345 ¹⁴C yrs.

DISCUSSION

Determination of the freshwater reservoir effect for individual stream systems is critical for environments where aquatic shells can be used for age control, yet accurate quantification of FRE values over the late Pleistocene and Holocene is not straightforward. Analyzing shells collected live from the modern system is important as these results can identify if there are reservoir effects today, and depending on the nature of sampling, may indicate if there are spatial differences in the reservoir effect along the reach of the stream, temporal differences over the time of collection, or if there are systematic differences between organisms due to diet, metabolic processes, or micro-habitat.

Average FRE_{modern} values of 518 ± 65, 640 ± 34, and 707 ± 76 ¹⁴C yrs determined for 2020, 2021, and 2022 clearly indicate that there is a ¹⁴C reservoir effect in Four Mile Creek. A key question is how spatially and temporally variable is this FRE for Four Mile Creek. Although the analysis of 11 samples over three years is not enough to answer this question, the data do

suggest that the FRE is variable over space and time. For example, the highest FRE_{modern} values for individual samples came from the bedrock reach of Four Mile Creek for all three years.

The determination of the temporal variability in the FRE of Four Mile Creek is more difficult to assess. Like all stream systems, variations in precipitation annually will influence the proportion of base flow to surface flow in streams and therefore stream alkalinity will change seasonally (Omernik and Griffith 1986). Over decadal time scales, intense summer droughts have the potential to significantly increase the FRE in Four Mile Creek. The differences in the FRE_{modern} values determined for 2020, 2021, and 2022 may be the result of seasonal differences in precipitation. However, the only way to determine the annual FRE and track variability over time is to isolate the shell growth for the given year and determine its ¹⁴C concentration. In this study we tried to target shell growth from the year collected by analyzing small *Corbicula fluminea* shells (10–20 mm) and selecting the outer section of the shell for analysis. But, the entirety of annual shell growth was not selected, nor can it be assured that all of the shell growth occurred during the given year. This is also true for the analysis of the Physidae, which are thought to have a lifespan of approximately one year (Wlodzimierz et al. 1989), as the entire shell was used for analysis.

Disparate FRE values among different taxa of aquatic mollusks in Four Mile Creek is possible as a result of differences in diet, micro-habitat, and perhaps as a consequence of physiological differences. However, no differences were identified among the two taxa analyzed in this study. More importantly, unlike terrestrial mollusks where there are differences in the ¹⁴C content between taxa due to dietary preferences (e.g., Pigati et al. 2010), there are no indications of different FRE values among aquatic mollusks that live in similar habitats, yet little work has been done to test this. Research is needed to determine if there are any broad differences identifiable among aquatic gastropods, fingernail clams (Sphaeriidae) and freshwater mussels (Unionida).

Important advantages to measuring the FRE values of aquatic mollusks collected live include the clear identification of modern reservoir effects, the assessment of potential spatial differences along reaches of the stream, temporal differences if the annual shell growth is isolated, and if there are any differences among taxa. Two key limitations, however, are anthropogenic modifications of the watershed and the impact of nuclear bomb testing (i.e., bomb spike) in the 1960s on ¹⁴C concentrations in organic or inorganic carbon pools in the watershed. Anthropogenic modifications for the Four Mile Creek watershed include the construction of a large (Acton Lake) and many small (e.g., retention ponds) reservoirs in the watershed, installation of numerous shallow drain pipes in floodplain soils, as well as extensive deforestation of the watershed (Rech et al. 2018). The impacts of these modifications on the relative proportion of base flow in Four Mile Creek and alkalinity are unknown. Nuclear bomb testing in the 1960s doubled the ¹⁴C concentration of the atmosphere (Povinec et al. 1986), preventing the accurate assessment of ¹⁴C reservoir effects. Although atmospheric ¹⁴C concentrations have returned to pre-bomb levels, elevated ¹⁴C concentrations are still within wood, the soil organic carbon pool, and various other carbon reservoirs in the watershed. The trend of increasing FRE values from 2020 to 2022 may be the result of reduced inputs of organic carbon influenced by nuclear bomb testing.

The determination of the ¹⁴C concentration of matched pairs of fossil aquatic mollusk shells and organic material allows researchers to omit the impact of extensive modifications of the watershed and the influence of nuclear bomb testing. Moreover, if sampling density is high enough, researchers may be able to identify changes in FRE values over time due to climate

change. This method of determining FRE_{fossil} values, however, also has several challenges. Key assumptions of using matched pairs to determine FRE values include (1) that the vegetation and aquatic mollusk stopped incorporating ^{14}C from the atmosphere and stream, respectively, at approximately the same time, (2) that there has been no reworking of the wood or shell material in the stream system, (3) that there is no secondary contamination influencing the organic or shell material, and (4) that the organic material is representative of the time of deposition and is not the result of reworking due to bioturbation or plant growth.

Twigs or seeds are ideal organic material to ^{14}C date as they represent new growth and are likely in equilibrium with the atmosphere when they enter the stream system. Unfortunately, these materials are rarely found in conjunction with aquatic mollusk shells in stream deposits. Wood, or charcoal, is more commonly found in association with mollusk shells, yet the inner wood of trees is not in equilibrium with the atmosphere and may be decades to centuries older than the shell. Both wood and shells can be reworked by streams. Some aquatic mollusk shells, for example the aquatic gastropod Physidae, have shells that are thin and fragile and are unlikely to survive reworking and transport in a stream system. Other aquatic gastropods, though, have shells that are quite thick and robust and could survive reworking. Finally, secondary contamination of geologic samples is always a concern. Fossil shells, especially those <20,000 years old, are fairly straightforward to chemically pretreat and are not likely to suffer from secondary contamination (Pigati et al. 2004, 2009; Rech et al. 2011). Organic material, however, can easily suffer from secondary contamination by organic acids that are not entirely removed during the chemical pretreatment.

The weighted mean FRE_{fossil} value of 1029 ± 345 ^{14}C yrs, determined here for Four Mile Creek on the basis of five matched pairs of organic material and shells, is the result of the freshwater reservoir effect, as well as all other errors associated with ^{14}C dating samples in the fossil record. As such, the calculated FRE value and error are likely higher than the true FRE value and error for Four Mile Creek.

Application: Four Mile Creek Case Study

To assess the potential utility of using aquatic mollusks for age control we ^{14}C dated and applied the weighted mean of FRE_{fossil} correction of 1029 ± 345 ^{14}C yrs to date three fluvial terraces of unknown age in the Four Mile Creek floodplain as case studies. We did not use the FRE_{modern} values to correct the shell ages as they may be influenced by the bomb effect and only three years of analyses do not incorporate the true variability associated with annual to decadal climate variability and its impact on stream flow and alkalinity.

The first fluvial terrace we dated was sampled at Trench 1 (Figure 2). The fluvial terrace is situated ~4 m above the modern stream level of Four Mile Creek. Soils on this terrace are fairly red and possess a well-developed argillic horizon (Figure 6a). After excavation, no material was apparent for age control with ^{14}C dating. After approximately one hour of searching for mollusk shells in the upper portion of the fluvial sands and gravels by two team members, one team member found a 3 mm *Pisidium compressum* shell. Sands and gravels were then placed in a 5-gallon bucket, brought back to the lab, and closely looked through. This resulted in the identification of a 5 mm *Valvata* sp. shell. These two shells were chemically pretreated and returned a ^{14}C age of $14,889 \pm 44$ BP. When the weighted mean of FRE_{fossil} of 1029 ± 345 ^{14}C yrs was applied and the age was calibrated, the 2- σ calibrated age range for the sample was 15,810–17,820 cal yrs BP (Table 3).

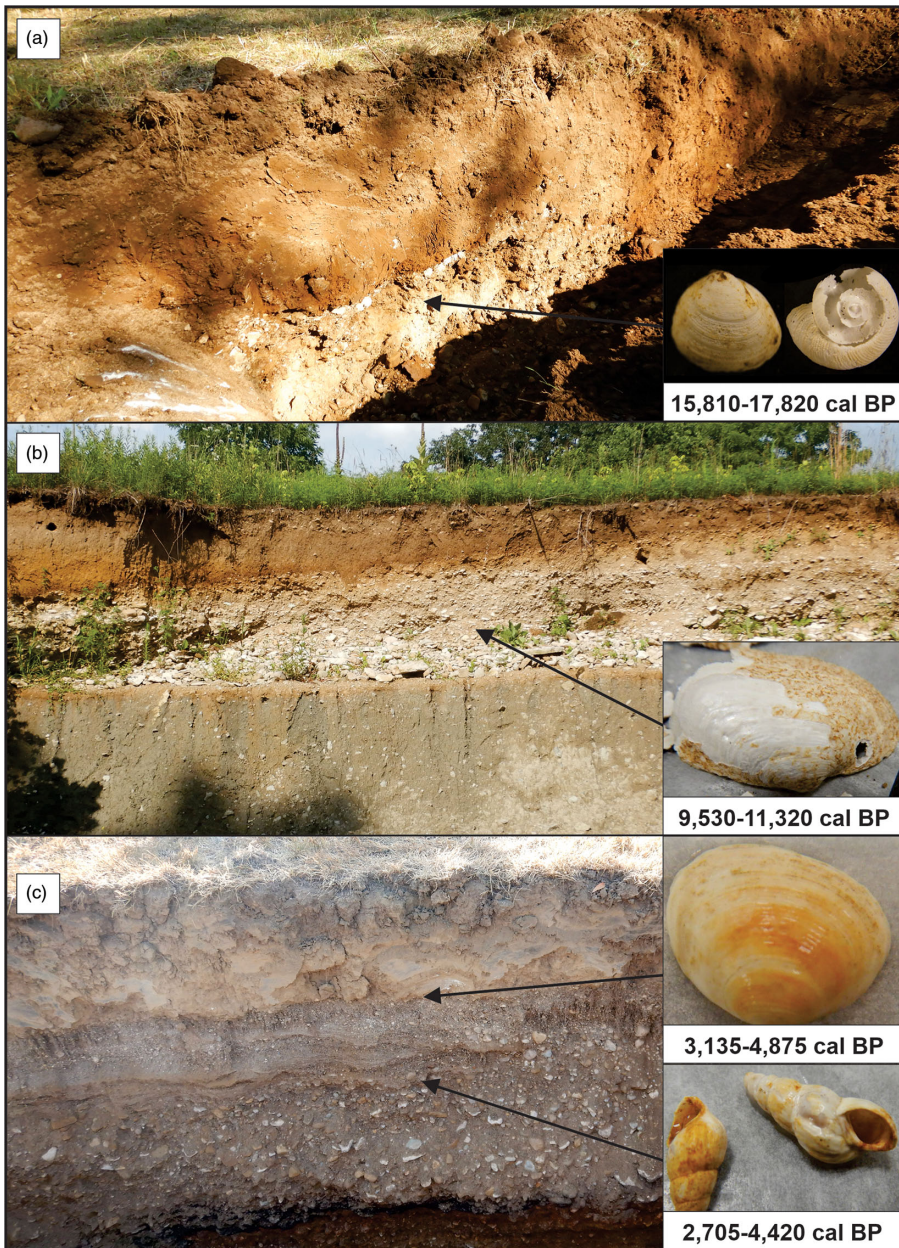


Figure 6 Examples of fluvial channel deposits with age ranges (2σ) for aquatic mollusks that have been reservoir-corrected and calibrated. (a) Trench 1, situated on a fluvial terrace ~ 4 m above modern stream level, with inset photographs of a *Pisidium compressum* shell (3 mm) and a *Valvata sp.* shell (5 mm). Shells were collected from fluvial channel deposits at a depth of -1.1 m and radiocarbon dated. (b) Station 2, situated on a fluvial terrace ~ 5.5 m above modern stream level, with inset photo of a Unionidae, tribe Anodontini, shell (46 mm). The shell was collected from a depth of -0.99 m from fluvial channel deposits and ^{14}C dated. Fluvial channel deposits overlie Wisconsin-aged glacial till ca. 24,000 years old. (c) Trench 6, situated on a fluvial terrace ~ 2.5 m above modern stream level, with inset photographs of *Sphaerium striatinum* (10 mm) collected from a depth of -0.84 m and two aquatic gastropods (*Elmia sp.*, 16 mm and 10 mm) collected from a depth of -1.6 m and ^{14}C dated.

Table 3 Radiocarbon ages of fossil aquatic mollusks utilized in the case study.

Sample ID	Lab #	Material	# of shells	Depth (cm)	¹⁴ C Age	¹⁴ C Age (FRE corrected)	1 σ	2 σ calibrated age range (cal yrs BP) [^]	P
Trench 1	D-AMS 044984	<i>Pisidium compressum</i> , <i>Valvata</i> sp. unidentifiable fragments	2+	–100 to –120	14,889 \pm 44	13,859	346	15,810– 17,820	1.000
Station 2	D-AMS 040993	Unionidae, tribe Anodontini	1	–99	10,254 \pm 35	9224	345	9530–11,320	1.000
Trench 6	D-AMS 040991	<i>Sphaerium striatinum</i>	1	–84	4681 \pm 23	3651	344	3135–4875	0.994
Trench 6	D-AMS 040992	<i>Elimia</i> sp. (<i>semicarinata</i> ?)	2	–155 to –173	4274 \pm 23	3244	344	2705–4420	0.998

[^] Age ranges are rounded to the nearest 10 years for ages >10ka and to the nearest 5 years for younger ages.

The second fluvial terrace of unknown age was a naturally exposed outcrop, Station 2, situated along Four Mile Creek (Figure 6b). Here, Wisconsin-aged glacial till (ca. 24 ka; Pigati et al. 2010) is exposed and overlain by fluvial gravels and overbank deposits (Figure 6b). A 4.6 cm freshwater mussel shell (Unionidae, tribe Anodontini), was collected from the outcrop at a depth of -99 cm, chemically pretreated, and ^{14}C dated to $10,254 \pm 35$ BP. This yielded a reservoir-corrected $2\text{-}\sigma$ calibrated age range of 9530–11,320 cal yrs BP.

Our final application was to date what was presumed to be a fairly young fluvial terrace based on its low elevation relative to the modern channel. Trench 6 is situated ~ 2.5 m above the modern channel. Excavation of a backhoe trench revealed a sequence of mostly channel sands and gravels with a thin unit (~ 25 cm) of overbank deposits on top (Figure 6c). Unlike the first two older sequences, this unit was extremely fossiliferous with the aquatic gastropod *Elmia* sp. being the most abundant. We ^{14}C dated two *Elmia* sp. gastropod shells from a depth of -155 to -173 cm and a small bivalve shell of *Sphaerium striatinum* at a depth of -84 cm. The *Elmia* sp. shells from a depth of -155 to -173 cm returned a ^{14}C age of 4274 ± 23 ^{14}C yrs and the *Sphaerium striatinum* shell from a depth of -84 cm yielded a ^{14}C age of 4681 ± 23 ^{14}C yrs. Once the weighted mean of $\text{FRE}_{\text{fossil}}$ correction was applied and ages were calibrated, the lower shells provided a $2\text{-}\sigma$ calibrated age range of 2705–4420 cal yrs BP and the upper shell provided an age range of 3135–4875 cal yrs BP.

The relative success of ^{14}C dating the aquatic mollusks from these three sites depends on the research questions and the availability of other material for age control. In the case of the two oldest fluvial terraces at Trench 1 and Station 2, there was no other material to ^{14}C date in the channel deposits and the ability to date the fluvial terraces with mollusk shells was a key advantage. The error associated with the FRE and calibration is large for ^{14}C dates of this age, but well within the error associated with most other Quaternary geochronologic techniques. The usefulness of ^{14}C dating aquatic mollusks is limited in its ability to assess the impact of short-duration climate events (e.g., Younger Dryas) on the fluvial system due to the margin of error.

In the case of the youngest, late Holocene fluvial terrace at Trench 6, the errors are large and they may limit the utility of the technique for dating relatively recent fluvial deposits. However, in this sequence no organic material was identified for ^{14}C dating. Moreover, within an adjacent trench (Trench 7) on the same fluvial terrace, organic material returned ages ~ 3000 years younger than shell ages (Table 2), suggesting that the organics suffered from secondary contamination, had been re-worked from above due to bioturbation, or were an old root. Even in cases of determining the age of late Holocene fluvial terraces, aquatic mollusks may be the only material to ^{14}C date. If organic material is present, it may be prudent to date both shell and organics as shells are less influenced by secondary contamination and reworking within the section from bioturbation. The shells can serve as a secondary check that the age of the organics is accurate.

CONCLUSIONS

Watersheds with carbonate bedrock in the midwestern United States provide for a diverse and abundant aquatic mollusk community as the high alkalinity waters allow these organisms to construct thick shells. Both of these factors are conducive to the incorporation of abundant mollusk shells into fluvial channel deposits, and the ability of some of these shells to survive weathering and diagenesis since at least the late Pleistocene. High stream alkalinity, however,

also leads to freshwater ¹⁴C reservoir effects that must be accounted for if these shells are used for ¹⁴C dating. In the midwestern United States, where thick accumulations of fluvial sediments occur within glacially excavated valleys, these shells are often the only material available for ¹⁴C dating. Thus, they provide the opportunity to date these deposits and tap these archives of climatic and environmental change.

This study identified the presence of freshwater ¹⁴C reservoir effects in the Four Mile Creek watershed of southwest Ohio both with the examination of specimens collected live and through match pairs of shell and organic material from the last 16,000 years. The average FRE_{modern} values were determined to be 518 ± 65 ¹⁴C yrs for 2020, 640 ± 34 ¹⁴C yrs for 2021, and 707 ± 76 for 2022, whereas the weighted mean of FRE_{fossil} value was determined to be 1029 ± 345 ¹⁴C yrs. Each method for assessing FRE values has advantages and disadvantages and should be used in conjunction to determine FRE values for a watershed. The use of matched pairs of shells and organic material from Quaternary deposits has many sources of potential error, besides just the FRE, and therefore is likely a conservative method for assessing FRE values.

As there has been no previous work on FRE values of aquatic mollusks in the midwestern United States, and relatively little work in other regions, there are many outstanding questions that need to be addressed. Two key questions include the potential impact of annual to decadal climate variations on the FRE value of streams as well as the magnitude of FRE values relative to stream discharge and the abundance of carbonate bedrock in a watershed. Future work needs to address these issues and potentially elucidate the spatial and temporal variability of FRE values to allow for the accurate dating of late Pleistocene and Holocene stream channel deposits.

SUPPLEMENTARY MATERIAL

To view supplementary material for this article, please visit <https://doi.org/10.1017/RDC.2023.93>

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