

1 **Review #1 author responses**

2 We thank the Reviewer for their broad and helpful analysis of this manuscript. It is clear the
3 Reviewer recognizes the extensive methodology and expertise that went into this study of why
4 certain groundwater discharge zones host repeated trout spawning, while a multitude of other
5 focused discharges within the same stream system do not. As you note, the inclusion of these
6 various interdisciplinary methods in one paper is challenging, and we needed to do a better job at
7 clarifying the underlying story. This starts with an updated title, for which we suggest:
8 “*Hydrogeochemical Controls on Brook Trout Spawning Habitat in a Coastal Stream*”. This new
9 title puts the emphasis on understanding the structural controls on why some discharges are
10 oxygen rich along this coastal stream, yet most are oxygen poor. Trout utilize the former for
11 spawning, as the eggs need dissolved oxygen to survive and properly develop. This relationship
12 between trout spawning and bed sediment oxygen is already known, and discussed in several of
13 the references we cite, so that point alone is not the focus of this research. In this study we could
14 have sampled the 40+ focused groundwater discharges (identified with fiber-optic heat tracing)
15 for parameters such as DO and EC, and compared to the n=3 that have been observed to
16 repeatedly host trout redds. That could likely have resulted in the kind of pore water chemical
17 dataset for which it would be appropriate to do various statistical analysis to indicate trout
18 chemical preferences in a more systematic way than we have done. Instead, which was not made
19 clear enough in the initial submitted text, we are operating under the assumption that for trout to
20 use groundwater discharge zones for spawning shallow pore water must be oxic.

21 We agree that revision of the main stated objectives is in order to best frame the motivation and
22 results of this study. These are now stated as:

- 23 *1. Identify repeat brook trout spawning locations, and determine if they are directly associated*
24 *with the preferential discharge of groundwater through interface sediments.*
- 25 *2. Develop a hydrogeochemical characterization of trout-preferred groundwater discharge zones*
26 *that can aid in their identification in other less-studied systems and potential inclusion in stream*
27 *habitat restoration efforts.*

28 As the Reviewer indicates, the different types of data collection used for this study were at times
29 hard to follow in the original version. We have responded with a restructuring of the manuscript
30 around themes, such as “*Spatial mapping of preferential groundwater discharges*” and
31 “*Visualizing streambed sediment geologic structure*”, and then divide the descriptions of the
32 specific methods and results under these themes. We feel this will help readers better follow the
33 main threads of the research presentation through the paper.

34 Main goals revolve around the use our unique multidisciplinary toolkit to understand why those
35 specific groundwaters have dissolved oxygen, and how discharge patterns develop at the meter-
36 scale. As the reviewer is likely aware, many statistically-based ecological studies result in
37 empirical relationships between fish and habitat attributes, but not necessarily process-based

understanding that can be readily transferable to other systems and future scenarios. We feel our study is valuable in showing trout in this coastal stream directly utilize discharges on meander bends that cut into mineral soils, as depicted in conceptual Figure 9. This conceptual understanding, supported by various data types and not known previous to this study, is powerful as it can directly be used to guide stream restoration and to identify possible spawning zones in other coastal streams using surficial soils maps and high-resolution elevation data such as LIDAR. Further, we show how the physical discharge of groundwater creates slumped alcoves outside of the main river flow that may offer additional favorable aspects for redd formation, such as reduced stream velocities.

The updated Table 1 is now reorganized by theme (eg streambed groundwater discharge zones vs near-bank spawn zones) and mean data are shown for each species. Newly added chemical and isotope data are shown in Table 2, and the vertical groundwater flux rates in Figure 6 have been converted to box plots so streambed seepage and spawn sites can be more easily compared.

We have now also put all data collection points on a common spatial reference scale: distance from an upstream bridge crossing/fish ladder. This is now reflected in all Figures, including Figure 4, which before had (in a confusing way) started with spatial reference at an arbitrary mid-stream point.

Specific comments

1. Line 47: Using the word “preferred” is problematic because this paper does not actually statistically analyze preference which would require an explicit comparison to what is available. I recommend removing the word “preferred” from the manuscript.

We agree. We now use the terminology “repeat spawning zones” to directly correspond with the annual observations of trout behavior (physical identification of spawning trout and geolocation of dropout PIT tags).

2. The manuscript refers to 10 years of observation that have gone into counting and mapping redds and spawning behavior, but these data (and sample sizes, etc.) are never presented. Perhaps the authors refer to a manuscript that published these data. In any case, it would be better to temper this kind of wording so that the readers won't be expecting some kind of explicit analysis of these data.

The wording in this section has been revised to explicitly state what type of trout spawning observations were made

3. Line 58: In many places, words and phrases are used that are colloquial and informal (e.g., short circuit, dropout, choked, etc.). Although I understand what the authors mean, many readers would not understand these phrases, so it would be best to go through the whole manuscript and eliminate all of this informal language.

We have attempted to revise the manuscript with an eye for informal language (eg “choked” was removed, among other phrases) but “short circuit discharge” and “dropout” PIT tags are known and accepted terms. For example short-circuit flowpaths are described in detail in the highly-cited Conant Jr. (2004) paper.

4. These are minor issues, but we refer to “coldwater and warmwater fishes” not “cold-water fish. It's just convention that this wording is recommended by the fisheries community (American Fisheries Society). In

this sense, it is different from “cold-water seep”, for example, which is a hydrologic rather than ecological feature. Also, please be careful about referring to refugia, which is the plural form of refuge or refugium. Thus, we don’t write “refugias”.
 Instances of “cold-water fish” and “refugia” have been revised
 5. Line 182: Shouldn’t this be in hectares? The units have been converted to hectares
 6. Line 193: The word “niche” does not have a scale per se, so it isn’t appropriate to use it this way. Be more explicit about what scale you are writing about. “niche” has been dropped here
 7. Line 236: No information is provided on the number of fish that were tagged. As outlined in my general comments, it would be better to keep the discussion of fish general. We have now tried to do that in the revised section regarding trout observations
 8. Line 343: Can you cite the previous work on the distribution of brook trout spawning? Otherwise, this can’t really be presented as a result because we have no data to evaluate in relation to this statement. This is just a summary statement for the findings of this work, not reference to previous work. We can see how this was confusing, and have deleted the statement.
 9. Figure 1b: This is really too small to examine and appreciate. It’s really impressive, but honestly the dots are too busy and crowded. We agree. The stream-scale FO-DTS data has now been broken out and enlarged in the new Figure 1
 10. Figure 7: The red and blue symbols are really hard to see on the graph because they overlay one another. Can you jitter them a little so that it is easier to see them? Yes, the point is that they directly overlay (similar chemistry), which of course make the background symbol harder to see. The background point is enlarged so that all 3 symbols are visible.

Review #2 author responses

The paper presents an extensive field data analyzed to identify the controls of preferential brook trout spawning sites. The data has not only been obtained for this particular study but represents a synthesis of newly acquired and existing data. However, the different data types as well as their location and timing of the collection make it hard to follow the story.
 Thank you for the time invested in this review. We agree that the original submitted manuscript was too hard to follow. Our group was so familiar with the site and study after several years of work that we did not appreciate how confusing it would be for the reader to piece together all of the various methods and results. With a fresh eye, and the helpful feedback from the first round of review, we have revised the title, main objectives, and overall organization of the paper. The “story” is now based around themes such as “*Spatial mapping of preferential groundwater discharges*” and “*Visualizing streambed sediment geologic structure*”, and then divide the descriptions of the specific methods and results under these themes. We feel this will help readers better follow the main threads of the research presentation through the paper.

My impression is that this is not because of the data as such but largely how it is presented. First I would have expected a rigorous evaluation of the factors that characterize the three preferred spawning sites such as EC, temperature and oxygen and GW discharge and how these conditions are different from the other GW discharge sites which are not used for spawning. However, there is no synthesizing figure or table or section where I can learn about this.

The original Table 1 showed the SpC and DO data from various spawn sites and sampled streambed groundwater discharge sites, but the data were organized by year of data collection instead of theme, making the Table difficult to interpret. The updated Table 1 is now reorganized by theme (eg streambed groundwater discharge zones vs near-bank spawn zones) and mean data are shown for each species. We have added new Table 2 that includes water quality data and stable water isotopes to further show distinguishing characteristics of repeat spawning zones.

The results section is mostly a description of the individual results of the different methods. The second part of the paper should then look into the regional geological setting that ultimately determines the conditions at the spawning sites. I suggest the restructure the paper, clearly following the objectives the authors state in their introduction. They provide an excellent guideline for the entire paper.

As mentioned above, the paper has been reorganized by theme instead of method, which makes for a better flow of the main story.

Additionally, many figures lack spatial reference or use non-unique references. For example in Fig. 4 and Fig. 7 the x-axis seem to start at different locations. Figure 5 has no x-axis at all. Please see also my specific comments below. Overall I found it very hard to keep track of which measurements have been conducted where and when.

We have now put all data collection points on a common spatial reference scale: distance from an upstream bridge crossing/fish ladder. This is now reflected in all Figures, including Figure 4, which before had (in a confusing way) started with spatial reference at an arbitrary mid-stream point.

Specific comments:

Figures: Generally, I recommend using consistent symbols and consistent spatial reference. Otherwise it is hard to recognize spatial setup. E.g.: in figure 6 spawn zones 1 2 3 are blue, red and green; figure 7: red, red blue; figure 8: blue green purple.

Figure 6 has been changed to box plots as recommended by the Reviewer. Figure 8 shows data at high spatial resolution in the shallow streambed, and the symbology was changed as to be less-confusing when following Figure 7.

Figure

1: Please indicate where b and c are located in a Figure 4: The decreasing order of the x-axis is confusing

The length of this FO-DTS deployment is now shown in Figure 2a, and the x-axis is now shown in total stream length from the common upstream point

Figure 5: x-axis missing, What is Loc 15?

Radar collection time is now shown on the x-axis. This cannot readily be converted to a linear spatial scale due to differential velocity of the river with distance, but the specific points of groundwater discharge zones of interest were directly marked in the record, as is now shown on the Figure.

Figure6: Would it

harm to show boxplots instead of the time series? The exact timing of the variations does not seem to play a role.

We have made this change

Figure 7: x-reference seems different from Figure 4 These Figures now have a common spatial reference

I.128: maybe explain which methods are lumped under the term 'geophysical remote sensing' I. 156-160: These objectives are absolutely reasonable but they are not reflected

in the structure of the results or the discussion section. Just an example: the discussion starts with heat tracing. Why? The paper has been reorganized by theme as discussed above
It should be about the Spawning sites I. 508:
The effects of shear stress and bed material seem to be important. So I wonder why this has been considered as factors controlling spawning site preference. Both can be measured. Shear stress was not measured or estimated here, so this statement was removed

Review #3 author responses

We would like to thank the Reviewer for the careful attention to our submitted manuscript, for the overall favorable impression of the work, and for the constructive suggestions. Through the combined 3 reviews we have identified several common themes that will improve the clarity and impact of this presentation. In regards to your 4 main points:

1. On the title, it sounded very exciting but it didn't come across to me why this is called "working backwards" after I read the entire manuscript multiple times. So I would suggest to either change the title or emphasize in the paper why this is considered "working backwards" compared to existing practices.

Our intention with the "working backwards from streambed thermal anomalies" title was to indicate how our work moves beyond measuring water fluxes and dissolved chemistry at the streambed interface, and into the source aquifer to develop a physical transport-based understanding of why certain groundwater discharge zones had favorable characteristics for trout spawning. In this case "moving backwards" is from a groundwater discharge flowpath perspective (moving upgradient from the discharge interface), but we realize this meaning is somewhat opaque. We suggest an updated title: "Hydrogeochemical Controls on Brook Trout Spawning Habitat in a Coastal Stream". This title plays on the strengths of this study, which are to illuminate how local river geomorphology, sediments, and flow groundwater flowpaths interact to generate oxygen-rich interface discharge zones in predictable positions along the reach.

2. It was very hard to follow all the data on a spatio-temporal map as they were taken at different places and at different times. It would be helpful to orient the readers where and when all those measurements were taken using a plot or table. It would be critical for this study to present the time window that is important for the fish habitat and whether the measurements were taken during that window. If they don't overlap, then explanations on why those measurements are relevant and useful would be needed.
All data have now been put on a common spatial scale (distance from upstream bridge/fish ladder), and all data collection sites are now shown in Figure 2. Measurements were all collected in summer, which should represent an end member for (higher) DO consumption in the streambed based on temperature-related kinetic rates. Extensive in-stream data collection is not possible during the spawning season as this would disturb the trout eggs in the shallow bed of this critical naturally reproducing (eg not stocked) trout system.

3. It was not clear when the drivepoint and minipoint samples were taken. If they were taken during the time when the fiber optic and other temperature profiles were deployed it would be helpful to indicate the sampling time on the temperature and seepage plots. The drivepoint and minipoint data were collected in 2014 and 2016 during the times of temperature data collection. All sampled locations are now noted on Figure 2a.

4. The impacts of hydrogeologic properties on the seepage and DO concentration could be demonstrated using a simple flow model, which could strengthen the paper substantially in linking all the valuable data together and generate useful insights. A simple reactive flow model could be generated to demonstrate uptake of oxygen around DOC sources (peat lenses) in the streambed, which differs from direct discharge from the mineral soil aquifer at meander bends. However, all three Reviewers commented on the sometimes overwhelming/confusing range of methods used here, and even the inclusion of a simple numerical model may not be a net positive to the manuscript readability. There are other empirical and model-based studies that have shown the relationship of DO uptake around in-situ DOC sources in sediment-water interface media, compared to DOC-poor media, and this referencing will be improved to support our interpretation.

A few other specific comments:

1. The locations of the fiber optic measurements are not found in Figure 2C. The spatial range of the FO-DTS measurements shown in Figure 4 are now shown in Figure 2.
2. most of the readers would need more information on how to read the GPR images. Yes, radargrams do need to be interpreted with some instruction/experience. We have highlighted the main reflectors (interpret as sand/peat interfaces) to help with this interpretation without adding extensive additional text
3. why there is no GPR information around spawn3? The instrument was not deployed in this area unfortunately
4. On Figure 6, not all lines show up on every plot, so more explanation is needed. The two legend boxes were meant to be one? x-axis with actual dates might work better for readers than the ordinal day. This has been changed to a box plot representation, as suggested by Reviewer #2
5. Figure 8, it might be helpful to plot 1-to-1 scatters for colocated specific conductivity and DO. We tried this, but the result was not clear.

~~Working backwards from streambed thermal anomalies: hydrogeologic controls on preferential brook trout spawning habitat in a coastal stream~~

Hydrogeochemical Controls on Brook Trout Spawning Habitat in a Coastal Stream

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~~Draft~~

~~Revised~~ manuscript prepared for submission to *Hydrology and Earth Systems Sciences*
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Abstract:

Brook trout (*Salvelinus fontinalis*) spawn in fall, and overwintering egg development can benefit from stable, relatively warm temperatures in groundwater seepage zones. However, eggs also are sensitive to dissolved oxygen concentration, which may be reduced in discharging groundwater- (i.e. seepage). We investigated a 2-km reach of the coastal Quashnet River, Cape Cod, Massachusetts, USA, to relate preferred fish spawning habitat to geology, geomorphology, and discharging groundwater dischargegeochemistry. Thermal reconnaissance methods were used to locate zones of rapid groundwater discharge, which were predominantly found along the center channel of a wider stream valley section. Pore-water chemistry and temporal vertical groundwater flux were measured at a subset of these zones during field campaigns over several seasons. Seepage zones in open valley sub-reaches generally showed suboxic conditions and higher dissolved solutes compared to the underlying glacial outwash aquifer. These discharge zones were cross-referenced with preferred brook trout redds, evaluated during 10 yr of observation, all of which were associated with discrete alcove features in steep cut banks where stream meander bends intersect the glacial valley walls. Seepage in these repeat spawning zones was generally stronger and more variable than open valley sites, with higher dissolved oxygen and reduced solute concentrations. The combined evidence indicates that regional groundwater discharge along the broader valley bottom is predominantly suboxic due to the influence of near-stream organic deposits; trout show no obvious preference for these zones when spawning. However, the meander bends that cut into sandy deposits near the valley walls generate strong,

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338 oxic seepage zones that are utilized routinely for redd construction and the overwintering of trout
339 eggs. Stable water isotopic data support the conclusion that repeat spawning zones located
340 directly on preferential discharges of more localized groundwater. In similar coastal systems
341 with extensive valley peat deposits, specific use of groundwater discharge points by brook trout
342 may be limited to morphologies such as cut banks where groundwater flowpaths ~~can short~~
343 ~~circuits~~ do not encounter substantial buried organic material and remain oxygen rich.

Introduction

The heat tracing of waters can be used to map a distribution of discrete groundwater discharge zones throughout surface water systems at times of contrast between surface and groundwater temperature. The measurement of water temperature from the reach to watershed scale is now possible using thermal infrared (TIR) and fiber optic distributed temperature sensing (FO-DTS) methodology (Hare et al., 2015). Remote TIR data collection throughout the river corridor has been enabled by handheld cameras, piloted aircraft, and the rapidly evolving capabilities of Unmanned Aircraft Systems. Researchers are capitalizing on the ongoing refinement of these technologies to identify zones of focused groundwater seepage to streams to map potential discrete preferential cold water fish habitat such as summer thermal refugia (Dugdale et al., 2015). However, surface thermal surveys alone do not directly indicate the suitability (e.g. dissolved gas and solute concentration) of discrete discharge aquatic habitat.

For example, dissolved oxygen (DO) concentration must be sufficiently high for cold groundwater seepage to provide support for fish life processes at the direct point of discharge to surface water (Ebersole et al., 2003), which is not apparent from thermal analysis alone. During summer warm periods in systems with suboxic groundwater, managed cold water fish species such as salmonids can face a tradeoff between occupying discrete zones of preferred water temperatures with near lethal DO levels, or stream sections that are too warm for long term survival (Mathews and Berg, 1997). The use of groundwater seepage zones as thermal refugia is further complicated by competition with aggressive invasive species (to the Northeastern USA) such as brown trout that compete with native trout for resources (Hitt et al., 2017). Streams at higher elevations may support reach scale cold water habitat where point scale thermal refugia are not needed under current climatic conditions, serving as vital “climate refugia” against rising

air temperatures (Isaak et al., 2015). In systems with reliably cold channel water in summer, which can also exist at low elevations when heavily influenced by discharging groundwater, salmonid fish may directly use groundwater seepage zones for spawning rather than thermal refuge.

Brook trout (*Salvelinus fontinalis*) are a species of char that are native to eastern North America, from Georgia to Quebec (MacCrimmon and Campbell, 1969). Populations have been stressed by warming temperatures and reduced water quality, particularly in low elevation areas (Hudy et al., 2008). Stream network scale tracking of fish has indicated brook trout directly utilize stream confluence mixing zones and groundwater seepage to survive warm summer periods (Baird and Krueger, 2003; Petty et al., 2012; Snook et al., 2016). Additionally, brook trout spawn in the fall, and eggs deposited in redds therefore develop over the winter before hatching in spring (Cunjak and Power, 1986). Oxygen use by the shallow buried embryos increases over the period of development (Crisp, 1981), and therefore DO concentration is a critical parameter of the pore waters in which the eggs are bathed. Several studies have demonstrated the importance of hyporheic downwelling in increasing shallow oxygen concentrations specifically at salmonid redds when streambed pore water is generally reduced in DO (e.g. Buffington and Tonina 2009; Cardenas et al., 2016). Fine sediments can reduce the efficacy of hyporheic DO exchange in spawn zones (Obruca and Hauer, 2016), and are actively cleared by trout during the spawning process (Montgomery et al., 1996).

The importance of hyporheic exchange to salmonid spawning may be limited in the lowland streams that are expected to harbor native cold water species in the 21st century: those with strong groundwater discharge. Groundwater seepage reduces the penetration of hyporheic flow from surface water (Cardenas and Wilson, 2006) and may shut down hyporheic flushing in

redds (Cardenas et al., 2016). Where hyporheic exchange does introduce oxygenated channel water into the shallow streambed, the downward advection of heat associated with near-freezing surface water in winter will also cool streambed sediments (Geist et al., 2002), potentially impairing egg development. Coaster brook trout, a life-history variant of native brook trout exhibiting potadromous migrations within the Great Lakes, have been shown to specifically prefer groundwater discharge zones for building redds (Grinsven et al., 2012). The development of trout in winter has been found to be positively correlated with warmer water temperatures as influenced by groundwater seepage (French et al., 2016), and therefore spatially discrete groundwater discharge zones with adequate DO may form preferred brook trout spawning habitat (Curry et al., 1995).

Multiscale physical and biogeochemical factors influence temperature and DO concentrations along groundwater flowpaths. In river valleys, discharge to surface water of locally recharged groundwater is expected to emanate from more shallow, lateral flowpaths controlled by local topography (Modica, 1999; Winter et al., 1998). Deeper regional discharge is expected to be more vertical through the streambed, as shown using our study site-specific topography in conceptual Figure 1a. Shallow groundwater flowpaths, particularly those within approximately 5 m of the land surface, will be more sensitive to annual air temperature patterns and longer term warming trends due to strong vertical conductive heat exchanges (Kurylyk et al., 2015). The distance of seeps from upgradient groundwater recharge zones will also affect seepage temperature dynamics and associated aquatic ecosystems due to future changes in temperature or precipitation (Burns et al., 2017). Therefore, working backwards from thermal anomalies into the landscape is critical to understanding the thermal stability of current and future point-scale preferential brook trout habitat (Briggs et al., 2017a). The complimentary

methodology of geophysical remote sensing, geochemical sampling, and vertical bed temperature time series can indicate the physical and chemical properties of groundwater flowpaths that source seepage zones utilized routinely by fish.

Coarse-grained mineral-dominated aquifers with little fine particulate organic matter and low dissolved organic carbon supply tend to result in generally oxic groundwater conditions (Back et al., 1993). The sandy surficial aquifer of Cape Cod, where our investigation took place, is a good example of a mineral soil-dominated flow system (Frimpter and Gay, 1979). Flow of groundwater through near-stream organic deposits, however, can result in inverted redox gradients toward the seepage interface, such that groundwater discharged to surface water is reduced in DO (Seitzinger et al., 2006). In sandy glacial terrain with superimposed peatland deposits, the specific flow patterns of groundwater to surface water in relation to buried peat will influence groundwater discharge biogeochemistry. Krause et al., (2013) found that streambed groundwater seepage was reduced in DO in zones with peat deposits, likely due to an increase in both near-stream residence time and localized source of dissolved organic carbon. Whether or not groundwater flowpaths are dominated by local or regional topography will influence where and how groundwater discharges to surface water, including possible contact with near-stream organic deposits (Figure 1a).

The heat tracing of waters can be used to map a distribution of spatially focused, or “preferential”, groundwater discharge zones throughout surface water systems at times of contrast between surface and groundwater temperature. The measurement of water temperature from the reach to watershed scale is now possible using thermal infrared and fiber-optic distributed temperature sensing (FO-DTS) methodology (Dugdale, 2016; Hare et al., 2015; Steel et al., 2017). Remote infrared data collection throughout the river corridor has been enabled by

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Coarse-grained mineral-dominated aquifers with little fine particulate organic matter and low dissolved organic carbon supply tend to result in generally oxic groundwater conditions (Back et al., 1993). The sandy surficial aquifer of Cape Cod, where our investigation took place, is a classic example of a mineral soil-dominated flow system (Frimpter and Gay, 1979). Flow of groundwater through near-stream organic deposits, however, can result in inverted redox

gradients toward the upwelling interface, such that groundwater discharged to surface water is reduced in DO (Seitzinger et al., 2006). In sandy glacial terrain with superimposed peatland deposits, the specific flow patterns of groundwater to surface water in relation to buried peat will influence groundwater discharge biogeochemistry. Krause et al. (2013) found that streambed groundwater seepage was strongly reduced in DO in zones with peat deposits, likely due to an increase in both near-stream residence time and localized source of dissolved organic carbon.

Interdisciplinary collaborations between physical and biological scientists are useful to better understand how cold-water species utilize groundwater ~~seepage~~discharge-influenced stream habitat, and the larger landscape-scale controls on ~~seepage-zone~~discharge characteristics. While previous hydrogeological research in the coastal stream used for this study had focused on locating and quantifying discrete groundwater discharge (e.g. “cold anomalies”, Hare et al., 2015; Rosenberry et al., 2016), here we endeavor to understand the hydraulic and biogeochemical controls on seepage zone distribution utilized directly by native brook trout. In this groundwater-dominated stream (e.g. likely climate refugia), brook trout do not need to occupy discrete inflows for summer thermal refugia, but do favor certain ~~seepage~~upwelling zones for fall spawning. We compare over a decade of visual survey and electronic fish PIT-tag dropout data regarding repeat brook trout ~~redds~~spawning locations to a comprehensive physical and chemical characterization of groundwater seepage zones across 2-km of stream to:

1. Identify ~~preferred~~repeat brook trout spawning locations, and determine if they are directly associated with the preferential discharge of groundwater ~~seepage, and identify common characteristics (e.g. temperature, dissolved constituents) between these zones~~through interface sediments.

2. Develop a ~~hydrogeological understanding~~hydrogeochemical characterization of trout-preferred groundwater discharge zones that can aid in their identification in other less-studied systems and potential inclusion in stream habitat restoration efforts.

Site Description and Previous ~~Ecologic and~~ Hydrogeologic Characterization

Cape Cod is a peninsula in southeastern coastal Massachusetts, USA, composed primarily of highly permeable unconsolidated glacial moraine and outwash deposits. The largest of the Cape Cod sole-source aquifers occupies a western (landward) section of the peninsula (~~LeBlanc et al., 1986~~)(LeBlanc et al., 1986), and is incised by several linear valleys that drain groundwater south to the Atlantic Ocean via baseflow-dominated streams (Figure 2a). Strong groundwater discharge to one such stream, the Quashnet River, supports a relatively stable flow regime that has averaged 0.49 ± 0.15 (SD) $\text{m}^3 \text{s}^{-1}$ from 1986-2015 (~~Rosenberry et al., 2016~~)(Rosenberry et al., 2016). The lower Quashnet River emerges from a narrow sand and gravel valley to a broader area with well-defined lateral floodplains. Historical cranberry farming practices, abandoned in the 1950s, have modified the stream corridor (~~Barlow and Hess, 1993~~)(Barlow and Hess, 1993). Primary modifications included straightening of the main channel (reducing natural sinuosity), installation of flood-control structures, incision of shallow groundwater drainage ditches in the lateral peatland floodplain, and widespread application of sand to the floodplain surface. The current bank-full width of the main channel averages approximately 4 m.

The Quashnet River has long been recognized as critical habitat for a naturally reproducing population of native sea-run brook trout (Mullan, 1958) ~~with a genetically distinct population (Annett et al., 2012). Efforts to restore trout habitat by the group Trout Unlimited and others have been ongoing for over 40 yr (Barlow and Hess, 1993). These efforts include the removal of~~

550 flood-control structures and planting of trees along the main channel, and addition of wood
551 structures to stabilize banks and provide cover from airborne predators. Further, the
552 Commonwealth of Massachusetts purchased 31 acres in 1956 and an additional 360 acres along
553 the lower Quashnet River in 1987 and 1988 to protect the area from development. (Mullan, 1958)
554 with a genetically distinct population (Annett et al., 2012). Efforts to restore trout habitat by the
555 group *Trout Unlimited* and others have been ongoing for over 40 yr (Barlow and Hess, 1993).
556 These efforts include the removal of flood-control structures and planting of trees along the main
557 channel, and addition of wood structures to stabilize banks and provide cover from airborne
558 predators. Further, the Commonwealth of Massachusetts purchased 12.5 hectares in 1956 and an
559 additional 146 hectares along the lower Quashnet River in 1987 and 1988 to protect the area
560 from development. The Massachusetts Division of Fisheries and Wildlife has been monitoring
561 trout populations since 1988 and movement since 2007.

562 Groundwater influence on stream temperature is pronounced, particularly over the 2-km
563 reach above the USGS gage, below which stream stage is tidally affected. Temperature
564 influences in summer include a general downstream cooling with distance toward the USGS
565 gage (Hare et al., 2015; Rosenberry et al., 2016). Ambient regional groundwater temperature is
566 approximately 11 °C (Briggs et al., 2014), and strong conductive and advective exchange with
567 the proximal aquifer maintains Quashnet River water temperature well below the lethal threshold
568 for brook trout (maximum weekly average temperature >23.3 °C, Wehrly et al., 2007).
569 Therefore niche-scale thermal refugia are not a current concern in this system, as the stream
570 supports system-scale cold-water habitat that is likely to persist into the future. In winter,
571 seepage zones can be located as relatively warm anomalies increasing and buffering surface
572 water temperatures from ambient atmospheric influence (Hare et al., 2015).

Previous work has measured relatively large net gains in streamflow over the lower Quashnet River (Barlow and Hess, 1993; Rosenberry et al., 2016), attributed to groundwater discharge through direct streambed seepage and harvesting of groundwater from the floodplain platform via relic agricultural drainage ditches. Repeat deployments of fiber optic temperature sensing (FO-DTS) cables along the thalweg streambed interface (June 2013, 2014) indicate the greatest density of focused seepage zones occurs along the broader valley area approximately 1 km upstream of the USGS gage; this zone coincides with the largest gains in net streamflow (Hare et al., 2015). Based on the streambed interface temperature data presented by Rosenberry et al. (2016), Figure 1b shows how temperature sensitive fiber optic cables have been used to pinpoint possible groundwater discharge zones based on anomalously cold mean temperature and/or reduced thermal variance. Focused evaluation of FO-DTS anomalies with physical seepage meters and vertical temperature profilers confirmed localized, meter scale seepage zonation along the streambed (Briggs et al., 2014; Irvine et al., 2016a), where discrete colder zones indicated through heat tracing showed approximately 5 times the groundwater discharge rate of adjacent sandy bed locations only meters away (Rosenberry et al., 2016). Active heating of wrapped FO-DTS cables deployed vertically within an open valley streambed seepage zone indicated true vertical flow to at least 0.6 m into the bed sediments (Briggs et al., 2016b), an expected characteristic of more regional groundwater discharge (Winter et al., 1998; Figure 1a), rather than that driven by topography local to the river. Hyporheic exchange in the lower Quashnet River system is superimposed on the general upward hydraulic gradient to the stream, and therefore reduced to a thin, shallow hyporheic exchange zone (e.g. < 0.1 m depth) along the thalweg by these competing pressures (Briggs et al., 2014; Rosenberry et al., 2016), as has been shown for similar systems (e.g. Cardenas and Wilson 2006).

Methods

A combination of fish tagging, geophysical surveys, and focused pore-water sampling was used to investigate the interplay between the locations of preferential brook trout spawning and the local hydrogeology. For consistency, we adopt the numerical naming convention of Rosenberry et al., (2016) for previously identified persistent streambed seepage zones as shown in Figure 1b. We also refer to the 3 sites of known repeated trout spawning activity as Spawn 1, 2, and 3, from upstream to downstream, respectively (Figure 2).

Brook trout spatial behavior

Groundwater influence on stream temperature is pronounced, particularly over the 2-km reach above the USGS gage (Briggs et al., 2018a), below which stream stage is tidally affected. Ambient regional groundwater temperature is approximately 11 °C (Briggs et al., 2014), and strong conductive and advective exchange with the proximal aquifer maintains surface water temperature well below the lethal threshold for brook trout (maximum weekly average temperature >23.3 °C, Wehrly et al. 2007). Therefore point-scale thermal refugia are not a current concern in this system, as the stream supports system-scale cold-water habitat that is likely to persist into the future and serve as warming “climate refugia” (Briggs et al., 2018a). In winter, seepage zones can be located as relatively warm anomalies increasing and buffering surface water temperatures from ambient atmospheric influence.

Previous work has measured relatively large net gains in streamflow over the lower Quashnet River (Barlow and Hess, 1993; Rosenberry et al., 2016), attributed to groundwater discharge through direct streambed seepage and harvesting of groundwater from the floodplain platform via relic agricultural drainage ditches. Deployments of fiber-optic temperature sensing (FO-DTS) cables along the thalweg streambed interface indicate the greatest density of focused seepage

zones occurs along the broader valley area approximately 1 km upstream of the USGS gage number 011058837; this zone coincides with the largest gains in net streamflow (Hare et al., 2015). Based on the streambed interface temperature data presented by Rosenberry et al. (2016), Figure 1 shows how temperature-sensitive fiber optic cables have been used to pinpoint possible groundwater discharge zones based on anomalously cold mean temperature and/or reduced thermal variance. Focused evaluation of FO-DTS anomalies with physical seepage meters and vertical temperature profilers confirmed localized, meter-scale seepage zonation along the streambed where discrete colder zones indicated through heat tracing showed approximately 5 times the groundwater discharge rate of adjacent sandy bed locations only meters away (Rosenberry et al., 2016). Active heating of wrapped FO-DTS cables deployed vertically within an open valley streambed seepage zone indicated true vertical flow to at least 0.6 m into the bed sediments (Briggs et al., 2016), an expected characteristic of more regional groundwater discharge (Winter et al., 1998), rather than that driven by valley topography local to the river. Hyporheic exchange in the lower Quashnet River system is superimposed on the general upward hydraulic gradient to the stream, and therefore reduced to a thin, shallow hyporheic exchange zone (e.g. < 0.1 m depth) along the thalweg by these competing pressures (Briggs et al., 2014), as has been simulated for similar stream systems (e.g. Cardenas and Wilson 2006).

Methods

A combination of fish tagging and visual spawning observations, heat tracing, geophysical surveys, and focused pore-water sampling was used to investigate the interplay between the locations of preferential brook trout spawning and the local hydrogeology. For consistency between varied methods and years of data collection, all sample locations are spatially referenced as downstream channel distance from the fish ladder river crossing at the

upper end of the study reach (Figure 2).

Observations regarding repeat spawning locations

Observations regarding discrete repeat brook trout spawning locations were made opportunistically as part of an ongoing PIT (Passive Integrated Transponder) tagging study of the native reproducing population of the Quashnet River. Large-scale trout movements are continuously monitored in the lower Quashnet River at 3 stationary fish counting sites (Figure 2a). However the spatial resolution of these counting sites, separated by hundreds of meters, is not adequate to study how brook trout utilize specific decimeter- to meter-scale groundwater discharge zones. For this finer scale characterization, dropped fish tags have also been located through roving surveys using a handheld portable PIT antenna (Biomark, Inc.) conducted in spring and fall since 2007. ~~In addition to tagged fish location at the time of these surveys, spawning brook trout were located visually during fall data collection events and clustering behavior captured within one seepage feature by underwater video in 2015 using a GoPro Hero camera (San Mateo, CA). Also the~~ The dropout of PIT tags from the fish body is a process that is more likely to happen during spawning behavior in salmonids (Meyer et al 2011), so dropped tags were located ~~and spatially mapped electronically and spatially mapped to reveal discrete zones of repeat spawning. Although these roving surveys do not yield the temporal continuity the instream counting gates, clustering of dropped tags can be mapped at the sub-meter scale, presumably directly at trout redds. In addition, spawning brook trout were located visually during annual fall data collection events by Massachusetts Fish and Wildlife Staff, with redd development behavior captured within one seepage feature by underwater video in 2015 using a GoPro Hero camera (San Mateo, CA). We refer to the 3 most prominent sites of brook trout spawning within the study reach as Spawn 1 (113 m), Spawn 2 (146 m), and Spawn 3 (2062 m).~~

from upstream to downstream, respectively (Figure 2).

Fiber optic distributed temperature sensing

Spatial mapping of preferential groundwater discharges

To augment ~~previously~~ existing streambed interface thermal surveys for preferential groundwater discharge (e.g. ~~Rosenberry et al., 2016; Figure 1b~~), ruggedized fiber-optic cables suitable for stream use were deployed along each bank from approximately 160 m upstream of the middle fish counter through the Spawn 3 meander bend for approximately 450 m total length (Figure 2a). (e.g. Rosenberry et al., 2016; Figure 1) and the bank-dependence of discharge location, ruggedized fiber-optic cables suitable for stream use were deployed in the river along the base of each bank from 1700 to 2160 m on June 10 through June 12, 2016 (Figure 2a). Two separate cables weighted with stainless steel armoring were installed directly along the foot of each bank on top of the streambed interface. Single-ended measurements made at the 1.01 m linear spatial sampling scale were integrated over 5-min intervals on each channel by an Oryx FO-DTS control unit (Sensornet Ltd.). During the same period, data were also collected along a high-resolution wrapped fiber-optic array for a dataset described in ~~Kurylyk et al. (2017)~~ Kurylyk et al. (2017) but not shown here; this experimental setup resulted in measurements for each channel of 4 instrument channels recorded at 20-min intervals. Calibration for dynamic instrument drift was performed automatically using an approximately 30-m length of cable for each channel submerged in a continuously mixed ice-bath and monitored with an independent Oryx T-100 thermistor.

Ground Penetrating Radar

—— Ground penetrating radar (GPR) has been successfully applied to several surface

water/groundwater exchange studies to characterize underlying peat and sandy deposits (e.g. Lowry et al. 2009; Comas et al. 2011) due to strong expected differences in matrix porosity (water content), which can exceed 70% in peat (Rezanezhad et al., 2016). An upstream to downstream GPR profile was collected on July 7, 2016, using a MALA HDR GX160 shielded antenna (MALA GPR, Sweden) hand-towed down the thalweg from a small inflatable watercraft. The locations of major seep and spawning sites were marked on the digital GPR record during data collection. The GPR data were processed using Reflexw software (Sandmeier, Germany) to convert reflection time to interface depth.

Temporal groundwater discharge characterization

Temporal patterns in vertical groundwater discharge flux rate can indicate source flowpath hydrodynamics, and can be derived from bed temperature time series, as reviewed by Rau et al., (2013). Custom “1DTempProfilers” designed specifically for the quantification of groundwater seepage (Briggs et al., 2014) were used to monitor streambed temperature over time along a shallow vertical profile. Profilers were deployed in zones of known focused groundwater discharge and/or preferential trout spawning from June 11 (day 162) to July 13 (day 193) in 2014; August 21 (day 233) to September 13 (day 247) in 2015; and June 5 (day 157) to July 9 (day 191) in 2016. Individual thermal data loggers (iButton-Thermochron DS1922L, Maxim Integrated) were waterproofed with silicone caulking and inserted horizontally into short slotted-steel pipes (0.025 m diameter). The shallow thermal profilers were driven vertically into the streambed so that sensors were positioned at some combination of 0.01, 0.04, 0.07, and 0.11 m depths. Data were collected at temporal intervals of 0.5 hr in 2014, 2015, and 1 hr in 2016. Rosenberry et al. (2016a) found that when a subset of the 2014 streambed temperature data presented here were analyzed using the diurnal signal amplitude attenuation models employed by

VFLUX2 (Irvine et al., 2015), a near 1:1 relation was found in comparison to physical seepage meter measurements of groundwater discharge ranging from 0.5 to 3 m d^{-1} . This strong relation was likely enabled by using in-situ measurements of thermal diffusivity (K_e) for modeling as suggested by Irvine et al. (2016) using the diurnal signal phase and amplitude relations presented by Luce et al. (2013). A sequential diurnal signal-based K_e evaluation to inform amplitude attenuation based analytical fluid flux modelling was used here, and this approach is described in detail by Irvine et al. (2016b).

Geochemical Quantification of vertical groundwater discharge rates

Once preferential discharge locations are located along the streambed with FO-DTS, actual vertical discharge rates can be assessed with a variety of methodologies (Kalbus et al., 2006). Temporal patterns in groundwater discharge flux rate can indicate source flowpath hydrodynamics, and be derived from bed temperature time series using vertical temperature signal transport characteristics, as reviewed by Rau et al., (2013). Custom “1DTempProfilers” designed specifically for the quantification of groundwater discharge (Briggs et al., 2014) were used to monitor streambed temperature over time along a shallow vertical profile. Profilers were deployed within a subset of the thermal anomalies previously identified with FO-DTS. The profiler deployment locations were chosen to represent a range of preferential groundwater discharge rates/characteristics based on the on the observed FO-DTS temperature anomalies, e.g. anomalies of varied mean temperature and buffering effect (Figure 1). These preferential groundwater discharges were located: 330, 880, 1045, 1070, 1410, 1470, and 2060 m approximate downstream distance from the fish ladder crossing. These groundwater discharge locations are referred to with the prefix “GW” followed by the meter mark for the remainder of the manuscript, such that major streambed seep 330 m downstream of the fish ladder is referred

to as “GW330”. Data were collected at various locations from June 11 to July 13 in 2014; August 21 to September 13 in 2015; and June 5 to July 9 in 2016. These deployments included installation of 1DTempProfilers at the nearbank and channel sides of observed repeat spawning zones.

Individual thermal data loggers (iButton Thermochron DS1922L, Maxim Integrated) were waterproofed with silicone caulking and inserted horizontally into short slotted-steel pipes (0.025 m diameter). The shallow thermal profilers were driven vertically into the streambed so that sensors were positioned at some combination of 0.01, 0.04, 0.07, and 0.11 m depths. Data were collected at temporal intervals of 0.5 hr in 2014, 2015, and 1 hr in 2016. Rosenberry et al. (2016a) found that when a subset of the 2014 streambed temperature data presented here were analyzed using the diurnal signal amplitude attenuation models employed by VFLUX2 (Irvine et al., 2015), a near 1:1 relation was found in comparison to physical seepage meter measurements of groundwater discharge ranging from 0.5 to 3 $\text{m}^3 \text{d}^{-1}$. A similar diurnal signal-based in-situ streambed thermal parameter estimation is used here.

Streambed groundwater discharge and spawning zone pore-water characterization

Subsurface water samples were collected for chemical analysis at ~~ten locations in the stream along the 2-km study reach using~~ 7 major open valley seepage locations and 3 repeat spawn locations. Geochemical data collection occurred in 2014 and 2016 along with the 1DTempProfiler deployments, while stable water isotope data were collection in August 2017. For geochemical sampling, 0.0095 m (nominal) stainless steel drivepoints ~~that had been~~ were inserted to depths of 0.3, 0.6, and/or 0.9 m. ~~A 2.4 m length of relatively gas-impermeable tubing (and Masterflex norprene size 15) Norprenetubing~~ was attached to the drivepoint ~~and a~~. A peristaltic pump was used to ~~pump groundwater~~ extract pore water samples until free of obvious

turbidity (typically requiring 3 min of pumping) after which the pumping rate was slowed and, the groundwater samples were collected by pumping into 60-mL HDPE syringe barrels. First an unfiltered sample for specific conductivity was pushed from the syringe into a 30-mL -HDPE Nalgene sample bottle. Second, a filtered sample for anion analysis was collected after attaching a 0.2- μ m pore size (25-mm diameter) Pall polyethersulfone filter to the syringe. Lastly, the pumping rate was slowed again and an overflow cup was attached to the norprene sample tubing and held upright until overflowing, at which point DO was measured by a field colorimetric test using the manufacturer's evacuated reagent vials, ~~which were snapped inside the overflow cup and then read on the field photometer~~ (Chemetrics V-2000). DO concentrations were read twice and the test repeated using an alternative vial kit if results were near the concentration range limit or out of range. The collected samples were kept cool and out of the light and analyzed for Cl^- upon return to the laboratory using standard ion chromatographic techniques.

~~Pore-water-~~In addition to the drivepoint samples-, pore water were also ~~were~~-collected in June 2016 from shallow depths ~~ranging between~~ 0.015, 0.04, 0.08 and 0.15 m below the streambed surface at ~~the same locations as the drivepoints~~GW1045 and Spawn 1, 2, 3 using ~~minipoint~~MINIPOINT samplers ~~(e.g. Harvey and Fuller 1998). These small volume water samples were collected at slow rates using 0.32-cm stainless steel tubes with slots of 0.01 m forming the screen 0.005 m behind a clamped tip. The sample tubes were pre-aligned for deployment at selected depths (0.015, 0.04, 0.08 and 0.15 m) by passing each tube through fittings that gripped the tubes in an acrylic disc that was lowered until the slotted ends of the sample tubes reached the desired depths.(e.g. Harvey and Fuller 1998).~~ Water was pumped simultaneously from all depths using a multi-head pump that withdrew small-volume samples (15 mL) at low flow rates (1.5 mL min^{-1}) to minimize disturbance of natural subsurface fluxes

and chemical gradients. Pumped lines terminated at press-on luer fittings that were pushed onto 0.2- μ m pore size (25-mm diameter) Pall polyethersulfone filters. Samples for specific conductivity were collected whereas filtered samples were collected for anions in prelabeled 20-mL LDPE plastic scintillation vials with Polyseal™ caps. Sample lines were then attached to overflow cups and dissolved oxygen concentrations were measured as described above.

During a follow-up field effort in August, 2017 streambed pore water samples were collected at the Spawn sites and at GW1045, GW1140 (approximately 70 m downstream of GW1070), and GW1470. Additionally, two large hillslope springs were identified along the edge of the riparian zone, upstream of Spawn 1, using a handheld infrared camera (FLIR T640, FLIR Systems, Inc.). These exposed springs were sampled to identify a localized hillslope groundwater signature that would not be impacted by valley-floor peat deposits. Samples were drawn from push-point piezometers installed from 0.2-0.44 m below the sediment interface, with deeper samples collected in the hillslope springs to avoid surface organic material. Pore water was evaluated for SpC, DO, and stable water isotopes. Isotope samples were analyzed by the U.S. Geological Survey Stable Isotope Laboratory using dual-inlet isotope-ratio mass spectrometry. A substantial fraction of regional Cape Cod shallow groundwater exchanges with the numerous groundwater flow through lakes as it discharges to the coast (Walter and Masterson, 2002). It is therefore assumed that the regional Cape Cod groundwater isotopic signature is likely to indicate evaporative processes (Leblanc et al., 2008), offering a contrasting signal from locally-recharged hillslope groundwaters (no substantial evaporation). Local deuterium excess of contemporary waters can indicate groundwater that has been influenced by evaporation in lakes, and is therefore in disequilibrium with local meteoric waters. Deuterium excess was determined here as: $d\text{-xs} = \delta^2\text{H} - 8 * \delta^{18}\text{O}$ (Dansgaard, 1964).

As mentioned previously, historic cranberry farming practices extensively modified the Quashnet River valley including the incision of drainage ditches into the floodplain. Some ditches extend from the valley wall to the main channel, whereas others are shorter or cut at angles. In addition to characterization of pore water, 34 major drainage ditches (observed flowing water) and a stream thalweg profile were spot checked for specific conductivity on June, 16 2014 (~~day 167~~), using the SmarTroll probe- (YSI). At a subset of these ditch locations, filtered grab samples were collected and analyzed in the laboratory for Cl⁻ in a similar manner as for the mini and drivepoint samples described above. In June 2016, the dataset was augmented for 5 ditch confluence locations upstream of Spawn 1. ~~Also in June 2016, a streambank piezometer was installed on the hillslope 2.1 m lateral to the Spawn 3 cut bank to a total depth of approximately 3 m and grab samples were collected after the well was cleared. A basic estimate of Darcy flux to Spawn 3 was made assuming a horizontal gradient, measured at 0.23 compared to stream stage on June, 5 2016, and estimated sand hydraulic conductivity of 10 m/d. Finally, for comparison to Quashnet River data the characteristic regional groundwater chemical signature of the upgradient groundwater aquifer was derived from Frimpter and Gay (1979) and LeBlanc (1984) for wells outside of known contaminant influence.~~

Visualizing streambed sediment geologic structure

Ground penetrating radar (GPR) has been successfully applied to several surface water/groundwater exchange studies to characterize underlying peat and sandy deposits (e.g. Lowry et al., 2009; Comas et al., 2011) due to strong expected differences in matrix porosity (water content), which can exceed 70% in peat (Rezanezhad et al., 2016). An upstream to downstream GPR profile was collected on July 7, 2016 using a MALA HDR GX160 shielded antenna (MALA GPR, Sweden) hand-towed down the stream center channel with a small

inflatable watercraft. The locations of major seep and spawning sites were specifically marked
on the digital GPR record during data collection. The GPR data were processed using Reflexw
software (Sandmeier, Germany) to convert reflection time to interface depth.

Results

~~Only 3 small alcoves along the 2-km reach were. The hydrogeochemical characterization~~
~~of observed to be consistently used for spawning by brook, repeat trout, all of which were~~
~~associated with meander bend cut banks. Heat tracing, geophysical, and chemical methods~~
~~indicate these spawning zones coincide with localized, oxide and other major streambed~~
groundwater discharge zones are contrasted below. Observations regarding repeat spawning
locations.

~~Brook trout spatial behavior~~

~~— Observations regarding repeat spawning locations~~

Out of the dozens of ~~focused seepage~~ preferential groundwater discharge zones
~~found~~ geolocated along the Quashnet River in this and previous work (e.g. Figure ~~1b~~ 1), brook
trout appear to consistently utilize only three ~~zones~~ discrete streambed locations for repeat
spawning activity. These locations coincide with steep cut banks where the river channel
approaches the sand and gravel valley wall (Figure 2b,c). Specifically, trout were found to
occupy small “scaloped” alcove bank features (Figure 3a) that may be formed by groundwater
sapping and subsequent slumping of sandy bank materials. In winter 2016, fresh slumping and
direct seepage from the newly exposed sand wall was observed at Spawn 3 (Figure 3c); a larger
slump event had filled approximately 1/3 of the scaloped alcove at Spawn 2 by June 2016.
Brook trout were observed clustered along the inner bank area at the Spawn 1 location in fall

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2015 (Figure 3d), and this spawning behavior was captured using underwater video (Supplemental Video S1).

Dropout PIT tags have been ~~located~~found repeatedly in each of the 3 preferential spawn zones. Seven dropout PIT tags were located in the Spawn 3 zone in March 2017, by far the most dropped tags found in any one location since the tracking program began in 2007. The only other obvious ~~persistent~~-scalped bank features along the 2 km study reach are located at ~~open valley seepage Locations 14/15~~GW1045 (Figure 3b), ~~where Location 14 is near the bank and 15 is in the thalweg~~. Compared to the trout spawning zone alcoves along the valley wall cutbanks (e.g. Figure 3a), this ~~strong~~-open valley seepage alcove was ~~choked~~overgrown with watercress and thick (tens of centimeters) loose deposits of organic material ~~and spawning trout have not been observed there~~.

Fiber optic distributed temperature sensing

— The FO-DTS cables Spatial mapping of preferential groundwater discharges

As shown in Figure 1, previously collected FO-DTS data was used to guide data collection at a subset of representative preferential streambed groundwater discharges. Additionally, paired FO-DTS cables were deployed at the base of both stream banks through a lower reach section in 2016 (Figure 2c) ~~show~~, revealing differing thermal anomaly patterns ~~of focused seepage zones indicated by persistent, cooler anomalies in Figure (Figure 4; (Briggs et al., 2017b). Briggs et al., 2018c). The cable along the downstream-right bank captures a large approximately 8-m-long cooler zone at Spawn 3 (Figure 4b), and this seepage signature is spatially reduced but visible along the opposing bank (Figure 4a). Other thermal anomalies observed along one bank show little or no signature along the other. ~~A short section of cable~~~~

(approximately 2 m) was deployed out of the water and over the fish counter apparatus, and data from this zone show diurnal changes in stream water temperature lag air diurnal changes by several hours (Figure 4b). Air temperature dropped noticeably over the final 1.5 d of deployment (day 162), and smaller cool anomalies that appeared on warm days are no longer captured by the streambed FO-DTS deployment, but the Spawn 3 signature is still visible along both cables.

Ground Penetrating Radar

The GPR data collected along the thalweg adjacent to Spawn 1 and 2 indicate a contiguous thin layer of material underlies the sandy streambed that may Quantification of vertical groundwater discharge rates. Ambient streambed temperature signal data can be peat deposited over deeper sands and gravels (Figure 5a) (Briggs et al., 2017b). The GPR profile through open valley seepage zone Locations 14/15 and 18 shows the strongest shallow reflectors of anywhere along the open valley section. These discontinuous interface structures are interpreted as layered sand, gravel, interspersed with thicker peat deposits (Figure 5b). Otherwise, discontinuous reflections indicative of sediment type interfaces of variable depth are observed near downstream open valley seepage zones where attenuated GPR signals indicate thick lenses of buried peat with high water content (Figure 5c).

Groundwater discharge characterization

Diurnal signal-based K_e measurements derived from 2 1D TempProfilers inserted in sandy thalweg used to measure streambed thermal conduction parameters (Luce et al., 2013), which is particularly important when applying heat-based methods to quantify upward vertical fluid flux (Rosenberry et al., 2016), compared to downward fluid flux models that generally show less sensitivity to streambed thermal parameters. Diurnal signal-based thermal diffusivity measurements derived from a pair of 1D TempProfilers inserted in sandy channel sediments for a month in 2014 have the same geometric mean value of $0.11 \text{ m}^2 \text{d}^{-1}$ (Briggs et al., 2018c), and this

value is used [here](#) to model vertical groundwater discharge for all locations and data collection periods (Briggs, 2017). Upward fluid flux modeling is particularly sensitive to sediment thermal parameters (Briggs et al., 2014), so reasonable upper and lower bounds of flux magnitude were estimated as ± 1 standard deviation of the sub-daily calculations of K_e ($n=732$), or a K_e range of $0.10\text{--}0.13\text{ m}^2\text{d}^{-1}$, which is the upper end of the general range observed for interface sediments (e.g. Rau et al 2012). This uncertainty in thermal parameters could be expected to generally shift the estimated flux values $\pm 0.2\text{ md}^{-1}$ when mean values range $0.5\text{--}1.0\text{ md}^{-1}$, and up to $\pm 0.5\text{ md}^{-1}$ for mean values of 3 md^{-1} (e.g. Spawn 3); however, these shifts do not impact the general pattern of temporal variability observed primarily at spawn zones.

(Briggs et al., 2018c). Sub-daily groundwater discharge fluxes evaluated over similar spring/early summer time periods in 2014 and 2016 show relatively stable patterns at open valley seepage zones, generally $<1\text{ md}^{-1}$ (Figure 6a,e). At Spawn 1 and 3, seepage is stronger (2 to 3.5 md^{-1}) and more variable than at open valley zones, with some apparent relation to variations in the stream water stage evaluated at the USGS gage (Figure 2a). The Darcy-based horizontal seepage estimate through the Spawn 3 bank, made using the bank piezometer, is 2.3 md^{-1} , which is similar to the temperature-based seepage rates at the Spawn 3 interface (Figure 6e), and indicates lateral discharge through the cut bank wall from a more localized groundwater flowpath (Figure 1a). The Spawn 2 zone shows a reduced and more stable discharge rate during summer 2016, and is likely impacted by a large bank slump into this zone that occurred during the winter of 2016, partially filling the alcove. Seepage patterns collected at Spawn 1 and 2 in late-summer 2015 show greater temporal stability, even though the stream stage at the downstream USGS gage showed substantial variation (Figure 6b). Discharge rates along the inner bank wall of the scalloped bank spawn zones were consistently higher than at bed

areas located just a few meters away toward the ~~thalweg (Figure 6a,b)-channel.~~

~~Geochemical~~Streambed groundwater discharge and spawning zone pore-water characterization

Based on previous characterization, the ~~regional~~Cape Cod sand and gravel aquifer generally has high DO concentrations (9 - 11 mg/L), relatively dilute specific conductance (SpC, 62 μ S/cm), and dilute chloride concentrations (Cl^- , 9.3 mg/L) at depths ranging between 12 and 20 m (Savoie et al., 2012). The groundwater that discharges to the Quashnet River, however, is often strongly variable in all three of these parameters (~~Harvey et al., 2017~~), but SpC and Cl^- are ~~used only to indicate aquifer flowpath properties and not suitable spawn habitat as their range is well within general brook trout tolerances.~~(Harvey et al., 2018). In June 2014, drivepoint data were primarily collected in -open valley seepage zones identified with FO-DTS; (Figure 1); these locations are ~~generally strongly~~ suboxic ~~or to~~ anoxic at 0.3 and 0.6 m streambed depths (Table 1). ~~The exception is Location 2~~Highest streambed seepage DO is found at GW330 in the tighter upstream valley section, ~~which has a DO concentration of~~ (4.6 mg/L at both depths,) and Spawn 3, where DO is 9.0 and 7.6 mg/L at 0.3 m and 0.6 m depths, respectively. (Table 1). SpC is also variable, but lowest and similar to the regional signal at ~~Location 2 and Spawn 3~~-GW330 and Spawn 3. Note, SpC and Cl^- are used here to indicate aquifer flowpath hydrogeochemical properties, and not suitable spawn habitat based on chemical concentration, as their range is well within general brook trout tolerances.

Drivepoint data collected at the 0.3 m depth in June 2016, primarily around spawn zones, generally show high DO and relatively low SpC at the interior of Spawn Zones 1 and 3 near the cut bank (Table 1). Data collected a few meters toward the ~~thalweg~~main channel from these near-bank spawn locations are reduced in DO with increased SpC, ~~in an apparent departure from the regional groundwater signal.~~ The Spawn 2 data were collected at the toe of the recent large

sediment slump that had partially filled the alcove, and DO data are suboxic at 0.3 m (3.9 mg/L) but more oxygen enriched at 0.9 m depth (7.2 mg/L) indicating the potential for shallow streambed respiration that removes oxygen from discharging groundwater flow paths (assuming vertical flow). Spawn Zones 1 and 3 are enriched and reduced in DO at the 0.9 m depth, respectively-slumped material. In contrast to the spawn zones, major open valley seepage Locations 14 (near scalloped bank, Figure 3b) and 15 (adjacent thalweg) are location GW1045 is nearly anoxic at all depths with SpC similar to the 2014 stream water profile grab samples (n=8, 101.4 +/- 1.7 $\mu\text{S/cm}$); little. Little difference was observed between near-bank and thalwegchannel positions; at GW1045 (both are suboxic) even though a large scalloped seepage bank feature was observed (Figure 3b).

The drainage-ditch grab samples generally show Cl^- concentrations that are lower than the average 2014 thalwegchannel grab samples (n=10, 19 +/- 0.4 mg/L), though the 2 most upstream ditches are similar to stream water, and 2 open valley ditches are appreciably higher in Cl^- (Figure 7a). Spawn Zones 1, 2, and 3 approximate the lowest Cl^- concentrations observed in drainage ditches, and Spawn 3 has a similar concentration to the adjacent 2016 streambank piezometer in both the 2014 and 2016 data. An analogous pattern is shown in the more widespread SpC data, with many drainage ditches and all spawn zones having concentrations around 60 $\mu\text{S/cm}$, but several ditches cluster around the stream water average or higher, particularly in the open valley area. Concentrations of DO at the drainage ditch confluences were highly variable, showing no pattern with channel distance, ranging 3.1-8.4 mg/L in June 2014.

The shallow, discrete interval shallow pore-water samples collected with the minipointMINIPOINT system show that streambed SpC is appreciably lower than stream water, even at the 0.02 m depth, at all near-bank spawn zones (Figure 8a). Conversely, the shallow

~~thalweg~~channel sediments at Spawn 1 and open valley seepage ~~Location 14/15~~GW1045 approximate the stream water value for SpC. DO is high and stable along the shallow profiles (to 0.14 m) at the interior of Spawn Zones 1 and 3, but suboxic at the Spawn 1 ~~thalweg~~channel ~~sample~~ and Spawn 2 zones, and essentially anoxic ~~at Location 14~~ along the ~~entire profile~~. ~~Thalweg seepage Location 15 shows~~bank at GW1045. Center channel pore water samples at GW1045 show moderate oxygen enrichment at 0.02 m (4.6 mg/L), which may result from hyporheic mixing ~~at, as deeper intervals along the 0.04-0.14 m depths that~~same profile are nearly anoxic.

Underwater video collected here in the fall of 2015 indicates Quashnet River brook trout clustered tightly around an approximate 1-m² bed area in Spawn 1 (Figure 3d, Video S1), directly at the base of the sandy cut bank. During the June 2016 collection of pore-water data, drivepoints were installed precisely in this area. Chemical analysis of 0.3 m depth pore water shows a strong gradient from the near-bank Spawn 1 zone to the outer alcove area, with specific conductance rising dramatically (70.6 to 143.9 μ S/cm) and DO falling (7.28 to 4.41 mg/L) ~~away from the bank~~ (Table 1). Spawn 3 shows a similar pattern from near-bank toward main channel (60.4 to 82.1 μ S/cm SpC; 9.11 to 1.76 mg/L DO), and Spawn 2, although complicated by the large slump during the previous winter, shows an increase in SpC from 70.6 to 139.3 μ S/cm from the inner to outer alcove. Conversely, pore water collected at 0.3, 0.6, and 0.9 m depths in the open valley seepage alcove at ~~Location 14/15~~GW1045 (pictured in Figure 3b) are ~~virtually functionally~~ anoxic with elevated SpC compared to inner spawn zones, and little gradient from bank to ~~thalweg~~. ~~Fine-scale shallow streambed minipoint data mirror these deeper samples (Figure 8)~~channel.

Pore water data collected in August 2017 indicate that all three Spawn sites are similar to

emergent hillslope springs, characterized by relatively high DO and low SpC compared to major open valley streambed seepage zones that are anoxic with higher SpC (Table 2). Additionally, the stable isotopic signatures of the hillslope and Spawn zones are similar, but contrasted by the lower deuterium excess metric determined for the open valley seepages. This indicates that groundwater discharging through the streambed away from the hillslope shows the evaporative signature of groundwater flow through lakes, and can therefore be considered regional discharge, compared to locally recharged hillslope groundwaters apparently favored by trout for spawning.

Visualizing streambed sediment geologic structure

Radar data were collected over most of the study reach length depicted in Figure 2a, and although spatial reference data were not collected for each sample point due to integrated Global Positioning System failure, Spawn and groundwater discharge zones of interest were precisely marked in the record (Figure 5). The GPR data collected along the thalweg adjacent to Spawn 1 and 2 indicate a contiguous thin layer of material underlies the sandy streambed that may be peat deposited over deeper sands and gravels (Figure 5a). The GPR profile through open valley groundwater discharge locations GW1045 and GW1070 show the strongest radar signal reflectors of anywhere along the open valley section (Figure 5b). These discontinuous geologic structures are interpreted as layered sand, gravel, interspersed with thicker peat deposits. Otherwise, discontinuous reflections indicative of sediment type-interfaces of variable depth are observed near downstream open valley seepage zones where strongly attenuated GPR signals indicate thick lenses of buried peat with high water content (Figure 5b,c).

Discussion

Heat tracing reconnaissance ~~technology~~ technologies, such as FO-DTS and ~~TIR~~ thermal infrared, offer an efficient means to ~~spatially~~ comprehensively characterize a subset of

~~focused~~preferential groundwater discharge points at the reach to watershed scale (e.g. Figure 4b). Using the groundwater-fed Quashnet River as an example, Rosenberry et al. (2016) showed that cold streambed interface anomalies in summer indeed corresponded to discrete zones of particularly high groundwater discharge through streambed sediments. This spatial characterization of discharge points alone is not sufficient to ~~fully understand~~characterize the physical and chemical drivers of niche critical cold-water habitat, but ~~it can~~ greatly focus investigation of the points of higher weighted influence on surface water efficiently guide additional data collection, as was done here. Compared to more ~~random~~randomly distributed streambed field parameter surveys, or larger spatial scale evaluations of net groundwater discharge made with differential gaging, comprehensive spatial mapping of groundwater discharges is a great advance in the context of understanding ~~point scale habitat. Here we have capitalized on previous FO-DTS data collection (Figure 1b) to locate dozens of seepage zones along a 2-km reach that could be assessed for temporal fluid flux dynamics and chemical characteristics using subsurface data collection. However, in fast flowing streams even a few meters wide,groundwater dependent ecosystems. However, in fast flowing streams, FO-DTS cable placement on the streambed will likely impact which specific groundwater discharge zones are captured with FO-DTS, as shown here by applying cables along opposite banks through the Spawn 3 area (Figure 4). The largest ~~discharges~~seepage zones may have a spatial footprint that encompasses the streambed area from bank to bank (e.g., the Spawn 3 cold anomaly), but a subset of more discrete seepage zones are bound to be missed with a single linear cable deployment. We did not capture Spawn Zones 1 and 2 in early FO-DTS field efforts (Figure 1), but fish tracking indicated their importance to trout spawning behavior. Therefore, in studies of niche stream habitat as influenced by preferential groundwater discharge, a combination of heat~~

tracing and biological observation may be needed to both identify major discharge points and discern which points are directly used by the biota of interest (e.g. brook trout).

In a study of the regional Cape Cod aquifer condition, Frimpter and Gay (1979) state that groundwater is typically near DO saturation, except downgradient of peat or river bottom sediments, where consumption of DO allows the mobilization of natural iron and manganese. Visible observations along the open valley section, in addition to streambed sediment coring (Briggs et al. 2014), revealed widespread coating of shallow streambed sediment grains with metal oxides, consistent with the conceptual model of organic material influence on near surface groundwater (Figure 1a). Aquifer recharge passing through upgradient groundwater flow-through kettle lakes (e.g. Stoliker et al., 2016) may also serve to decrease the DO content of regional flowpaths that discharge vertically through the bed of the Quashnet River, although we hypothesize that localized peat deposits may be the primary control on groundwater discharge zone distribution and chemistry.

In a study of the regional Cape Cod aquifer condition, Frimpter and Gay (1979) state that groundwater is typically near DO saturation, except downgradient of peat or river bottom sediments, where consumption of DO allows the mobilization of natural iron and manganese. Visible observations along the open valley section, in addition to streambed sediment coring (Briggs et al. 2014), revealed widespread coating of shallow streambed sediment grains with metal oxides, consistent with the conceptual model of organic material influence on near-surface groundwater (Figure 9). Aquifer recharge passing through upgradient groundwater flow-through kettle lakes (e.g. Stoliker *et al.* 2016) may also serve to decrease the DO content of regional flowpaths that discharge vertically through the bed of the Quashnet River, although we hypothesize that localized peat deposits may be the primary control on both seepage zone

distribution and chemistry.

Out of the dozens of ~~focused seepage-preferential groundwater discharge~~ zones located along the lower Quashnet with heat tracing, most were suboxic to anoxic (~~Tables~~Table 1,2).

Brook trout ~~seem to~~ consistently prefer 3three areas for fall spawning, all along meander bend cut banks into the sand and gravel valley wall. Zones of locally enhanced seepage, likely controlled by subtle differences in sediment hydraulic conductivity, can lead to groundwater sapping of fines, reduction in bank stability, and consequent slumping of bank material into the river; this process was observed in real-time at the Spawn 3 meander in February 2016 (Figure 3c). Slumping effectively forms *seepage-driven* alcoves outside of the main flow ~~along banks~~ ~~where bed shear stress is reduced~~ and more suitable for redd placement, along with a more favorable coarse sand and gravel substrate (~~Bowerman et al., 2014; Hausle and Coble, 1976; Raleigh, 1982~~)(Bowerman et al., 2014; Hausle and Coble, 1976; Raleigh, 1982).

In other systems, trout have been observed to occupy microhabitat around and within groundwater discharge zones, even segregating by fish size and desirable temperature range (e.g., Figure 2.4.1.2 in Torgersen et al. 2012). ~~Here real-time observation and visual imagery show trout clustering tightly against the bank in Spawn 3 (Figure 3d, Video S1), where pore water was found to be more oxygen-rich and lower in SpC. Month-long time-series of bed temperature-derived fluid flux show that the vertical groundwater discharge rate is reduced considerably from inner to outer alcove zones, indicating a strong reduction in hydraulic gradient and/or decrease in effective streambed hydraulic conductivity. The evidence of higher near-bank vertical groundwater flux rates and DO, combined with lower SpC, indicates limited interaction between shallow groundwater flowpaths and peat against the meander bend cut banks, resulting in groundwater discharge zones most preferred by brook trout for spawning. The remote sensing~~

of streambed material with GPR indicates a relatively thin layer of streambed peat in the Spawn 1 and 2 thalweg area compared to open valley seepage zones (Figures 1 and 5). Therefore, it appears that even short travel distances through organic deposits toward the center channel at Spawn 1 and 2 may be sufficient to increase total dissolved solids and deplete DO, as observed in other systems (e.g. Levy et al., 2016), and render seepage zones undesirable for redd construction. This characterization is consistent with previous GPR data collected several kilometers upstream in a broad valley area in a study to assess possible naturalized channel form restoration (personal communication Maggie Payne, John Cody, Melissa Kenefick, Natural Resources Conservation Service, November 30, 2015). Their assessment found peat deposits >5 m thick in the central area of the valley, pinching out against the sands and gravels of the valley walls. Cored peat sections were indicative of a buried cedar swamp, which is typical of similar glacial depressions in the area (Hare et al., 2017). Here, real-time observation and visual imagery show trout clustering tightly against the bank in Spawn 3 (Figure 3d, Video S1) where pore water was found to be more oxygen rich and lower in SpC. Month-long time series of vertical groundwater discharge rates are reduced considerably from near-bank to near-channel at all spawning zones (Figure 6), indicating in part a reduction in streambed hydraulic conductivity as influenced by peat deposits under the main channel as observed in GPR data (Figure 5). The combined evidence of higher near-bank vertical groundwater flux rates and DO, combined with lower SpC, indicates limited interaction between shallow groundwater flowpaths and peat against the meander bend cut banks. It appears that even short travel distances through organic deposits toward the center channel at Spawn 1 and 2 may be sufficient to increase total dissolved solids and deplete DO, as observed in other systems (e.g. Levy et al., 2016), and render upwelling zones undesirable for redd construction. Therefore, near-surface channel sediments

may need to be specifically characterized in preferential groundwater discharge zones, as net chemical reactivity over the last ~1 m of transport may dominate that along km of upgradient groundwater flow through mineral soils.

Only where seepage was observed to emanate directly from the valley wall sands and gravels, such as the newly exposed slump in Figure 3C, may groundwater discharge reliably support overwinter trout egg development. These features are apparently similar to the numerous cold-water alcove patches observed ~~by Ebersole et al. (2003)~~in another stream system by Ebersole et al. (2003). In that study of preferential salmonid habitat, alcoves were often located where streams converged on valley walls and were the most abundant type of discrete cold-water habitat type identified. Conversely, valley wall alcoves were the least-common type of seep morphology observed along the Quashnet River. It is likely that the artificial reduction in channel sinuosity along the Quashnet River ~~through~~by farming practices has reduced possible higher-quality spawning locations ~~by focusing river flow away from the valley walls and oxide groundwater discharge.~~

Other bank alcove features with strong groundwater discharge found along the open valley section (Figure 3b) were highly influenced by organic material deposition and did not apparently support spawning habitat. Our research indicates that in lowland systems with organic-rich floodplain sediments ~~(e.g. Figure 1a), valley wall alcoves create favored brook trout spawning habitat via mineral soil-dominated groundwater discharge flowpaths. It seems reasonable to infer that these features would also create preferential thermal refugia in streams at times when main channel water exceeds fish thermal tolerances. valley wall alcoves alone create favored brook trout spawning habitat via local mineral soil-dominated groundwater discharge flowpaths as shown in conceptual Figure 9. This finding might help inform future ecologically-~~

based stream restoration practices in using the natural landscape to predict desirable preferential groundwater discharge points, as was recently done by Hare et al., (2017) to inform the engineering of a large-scale cranberry bog restoration.

The pore-water ~~chemical~~ SpC, Cl-, and DO data alone do not definitively show that seepage at the cut bank spawn sites is derived from more localized groundwater recharge, as opposed to regional groundwater that is unadulterated by buried peat lenses. However, the hydrodynamic data derived from long-term vertical temperature profiling in seepage zones does offer additional insight. In general, groundwater discharge rates are more variable at cut bank spawn zones than in the open valley streambed zones (Figure ~~6a,c~~ 6a,e), and this variability may be tied to shorter-term changes in local river stage and/or water table depth, impacting the hydraulic gradient. ~~The temporally consistent patterns of open valley discharge may be controlled by the regional gradient where the flowpath length term dominates, rendering the Darcy gradient relatively insensitive to discharge zone changes in river stage or bank proximity. Previous active streambed heating experiments have indicated open valley seepage to be vertical in nature to >0.6 m depth also indicating regional discharge (Briggs et al., 2016a), compared to lateral local discharge through the steep cut bank, indicated by the bank piezometer stream stage lateral gradient at Spawn 3-local hydraulic gradient. The relatively stable patterns of open valley groundwater discharge may be controlled by the regional gradient where the flowpath-length term dominates the Darcy relation, and is therefore relatively insensitive to local changes in river stage and water table fluctuations. Further, the stable water isotope data display evaporative signatures at the open valley streambed discharge sites, indicating regional groundwater that has passed through one or more upgradient flow-through lakes (Table 2). In contrast, the Spawn sites all show isotope signals that fall along the local meteoric water line, and therefore likely~~

represent recharge to the hillslopes more local to the river. These localized groundwater flow systems would be expected to be less-influenced by regional groundwater contamination, which is widespread in the regional Cape Cod aquifer (Walter and Masterson, 2002).

Groundwater drainage-ditch data collected along the river corridor indicate low SpC/Cl⁻ conditions exist for the majority of ditches throughout the lower Quashnet River riparian areas (Figure 7). The hillslope piezometer in sand and gravel at the down valley wall has a similar chemical signature along with high DO. This similarity is further indication that low-SpC groundwater discharges even to the lower portion of the river corridor, but is predominantly modified chemically by travel through near-stream organics. The relic drainage ditches allow discharging groundwater to effectively short circuit the valley floor peat deposits and remain high in DO, similar to the natural valley wall ~~cut bank alcoves. Future restoration strategies in similar systems~~ springs and cut bank alcoves. Future restoration strategies that seek to actively enhance groundwater discharge (e.g. Kurylyk et al., 2015a) may consider capitalizing on this short circuit behavior, possibly by auguring through buried streambed peat or movement of the stream channel toward the valley wall to create more desirable brook trout aquatic habitat.

Conclusions

The three ~~preferential spawn zones repeatedly utilized discrete spawning zone locations~~ that have been identified over 10 yr+ of observation ~~in the 2 km study reach~~ have coupled strongly discharging groundwater with high DO concentration. ~~The~~ A conceptual diagram of the hydrogeochemical setting of spawn zones vs other non-favored streambed groundwater discharge locations is shown in Figure 9. Spawn zones are located exclusively in side alcoves of the channel created by bank slumps along meanders where the river cuts into steep hillslopes

1168 along the glacial sands and gravel valley wall (Figure 1a). In the alcoves at the base of the cut
1169 banks, hillslope groundwater with high DO concentration is discharged through the streambed
1170 without appreciable loss of oxygen. Just a few meters away toward the main channel, however,
1171 groundwater consistently discharges at lower rates, is reduced in DO, and increased in SpC. The
1172 lowest oxygen concentrations in groundwater are associated with water emerging from the
1173 streambed adjacent to wide riparian areas that flank the Quashnet in the open valley section of
1174 the study reach, even though groundwater discharge rates were also relatively high. In the open
1175 valley ~~zone~~, where the stream is not near the valley walls, proximity to the stream bank does not
1176 seem to control seepage chemistry, and GPR data indicated thick zones of discontinuous
1177 streambed peat. In this and other groundwater-dominated streams that are expected to serve as
1178 climate refugia for future native trout populations, hyporheic exchange will be limited by strong
1179 upward hydraulic ~~gradients, and gradient~~. Therefore, preferential spawning habitat in such
1180 lowland valley systems may be primarily supported by discrete zones of oxic groundwater
1181 seepage upwelling at the meter to sub-meter scale as has been indicated by previous work ~~(e.g.~~
1182 Curry et al., 1995) (e.g. Curry et al., 1995).

1183 ~~In systems where all groundwater discharge is anoxic, preferential salmonid spawning~~
1184 ~~zonation may be controlled by points of downwelling hyporheic water where shallow sediments~~
1185 ~~remain high in DO (Buffington and Tonina, 2009; Cardenas et al., 2016). However, these~~
1186 ~~hyporheic areas will deliver cold surface water to shallow sediments during winter, which may~~
1187 ~~impair overwintering brook trout eggs (French et al., 2016). Here, and in many other coastal~~
1188 ~~systems, groundwater temperature is expected to range approximately 10–12 °C, which is an~~
1189 ~~ideal range for egg development (Raleigh, 1982). Points of oxic groundwater seepage devoid of~~
1190 ~~near-stream buried organics, combined with a recirculating side alcove and favorable sand and~~

~~gravel sediments, may provide ideal and unique groundwater seepage-enabled preferential spawning habitat for native trout.~~

In systems where all groundwater discharge is universally anoxic, preferential salmonid spawning zonation may be controlled by points of downwelling hyporheic water where shallow sediments remain high in DO (Buffington and Tonina, 2009; Cardenas et al., 2016). However, these hyporheic areas will deliver cold surface water to shallow sediments during winter, which may impair overwintering brook trout eggs (French et al., 2016). Here, and in many other coastal systems, groundwater temperature is expected to range approximately 10-12 °C, which is an ideal range for brook trout egg development (Raleigh, 1982). Points of oxic groundwater upwelling devoid of near-stream buried organics, combined with a recirculating side alcove and favorable sand and gravel sediments, may provide ideal and unique groundwater seepage-enabled preferential spawning habitat for native trout.

Stream surface or streambed interface heat tracing of groundwater discharge offers an efficient means to locate discrete seepage zones, but offers only limited insight into ~~the source~~ groundwater flowpath hydraulics and ~~biogeochemistry that impact localized trout habitat-geochemistry~~. A combined toolkit that also includes spatially-informed (using heat tracing) geochemical and ~~valley-scale geomorphic assessment may~~ isotope sampling and geophysical imaging can be ~~needed~~ used to ~~locate probable preferential seepage zones in other glacial systems, and guide stream ecological restoration design. A~~ trace groundwater flowpaths back into the source aquifer, and develop a robust hydrogeochemical characterization. Additionally, as digital elevation models become more refined and ~~are~~ combined with infrared data derived from Unmanned Aircraft Systems, remote identification of relatively small features such as the seepage alcoves described here should be possible. Comprehensive and process-

1214 based characterization niche stream habitat can be used to guide stream ecological restoration
1215 design that directly incorporates the local preferential groundwater discharge template.

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1449 **Tables**

1450 Table 1. 2014 and 2016 drivepoint pore-water chemistry data collected in major streambed
1451 groundwater ~~seepage~~discharge zones located with fiber-optic heat tracing, and in zones of
1452 observed repeat trout spawning directly along the bank (“~~in~~”) and farther toward the stream
1453 ~~thalweg~~ (“~~out~~”)-center channel.

<i>June 2014</i> <i>Location</i> <u>open valley</u> <i>groundwater</i> <i>discharges</i>	<i>0.3 m depth</i>		<i>0.6 m depth</i>	
	DO	SpC	DO	SpC
	mg/L	µS/cm	mg/L	µS/cm
<u>Location 2</u> <u>GW330</u>	4.6	53.8	4.6	61.3
<u>Location 4</u> <u>GW880</u>	1.4	97.7	3.4	65.1
<u>Location 15</u> <u>GW1045</u>	0.1	78.8	0.0	82.5
<u>GW1045 (bank)</u>	<u>0.16</u>	<u>105.5</u>	<u>0.39</u>	<u>104.0</u>
<u>GW1045 (channel)</u>	<u>0.31</u>	<u>99.1</u>	<u>0.18</u>	<u>96.4</u>
<u>Location 18</u> <u>GW1070</u>	0.2	100.0	0.2	89.8
<u>Location 21</u> <u>GW1410</u>	0.0	77.7	0.0	79.0
<u>Location 24</u> <u>GW1470</u>	0.1	69.1	0.0	64.3
<u>Location 27</u> <u>GW2060</u>	1.4	75.0	0.5	79.4
<u>Spawn 3 in</u> <u>mean</u>	<u>0.9.0</u>	<u>84.156.4</u>	<u>7.61.0</u>	<u>60.980.2</u>
<i>June 2016</i> <i>Location</i> <u>spawn</u> <i>ing locations</i> <i>(channel)</i>	<i>0.3 m depth</i>		<i>0.9 m depth</i>	
<u>Spawn 1 in</u>	<u>7.28</u>	<u>70.6</u>	<u>9.76</u>	<u>55.9</u>
<u>Spawn 1</u> <u>outchannel</u>	4.41	143.9	5.68	143.2
<u>Spawn 2 in</u>	<u>3.89</u>	<u>70.8</u>	<u>7.17</u>	<u>57.6</u>
<u>Spawn 2</u> <u>outchannel</u>	5.25	139.3	n/a	n/a
<u>Spawn 3 in</u>	<u>9.11</u>	<u>60.4</u>	<u>4.91</u>	<u>71.9</u>
<u>Spawn 3</u> <u>outchannel</u>	1.76	82.1	2.68	79.9
<u>mean</u>	<u>3.8</u>	<u>121.8</u>	<u>4.2</u>	<u>111.6</u>
<u>Location 15</u> <u>in</u> <u>spawning</u> <u>locations</u>	<u>0.463 m depth</u>		<u>0.399 m depth</u>	
		<u>105.5</u>		<u>104.0</u>

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<i>(bank)</i>				
Spawn 1 bank	<u>7.28</u>	<u>70.6</u>	<u>9.76</u>	<u>55.9</u>
Spawn 2 bank	<u>3.89</u>	<u>70.8</u>	<u>7.17</u>	<u>57.6</u>
Spawn 3 bank	<u>9.11</u>	<u>60.4</u>	<u>4.91</u>	<u>71.9</u>
(2016)				
Location 15 outSpawn 3			<i>7.6 (0.186</i>	
bank (2014)	<i>9.031</i>	<i>56.4991</i>	<i>m)</i>	<i>60.9 (0.6 m) 96.4</i>
mean	<u>7.3</u>	<u>64.6</u>	<u>7.4</u>	<u>61.6</u>

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Figure List

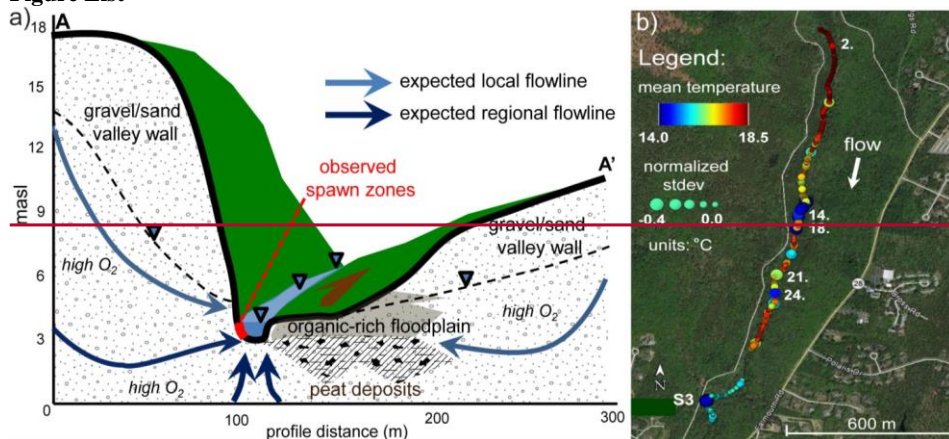


Figure 1. Based on previously published USGS work (e.g. Modica, 1999; Winter et al., 1998), the conceptual model in panel a) displays how groundwater discharge to lowland streams is expected to include locally sourced lateral groundwater discharge through valley wall features and more regionally sourced groundwater discharge vertically through the streambed. The topographic profile shown here (A-A') is derived directly from lidar data in the vicinity of observed preferential brook trout spawning habitat shown in Figure 2. In contrast to the sand and gravel valley walls, multiple methodologies used for this study indicate wider valley zone sediments to be rich in organic material, including buried peat deposits, consistent with known regional geology. Modified in format from

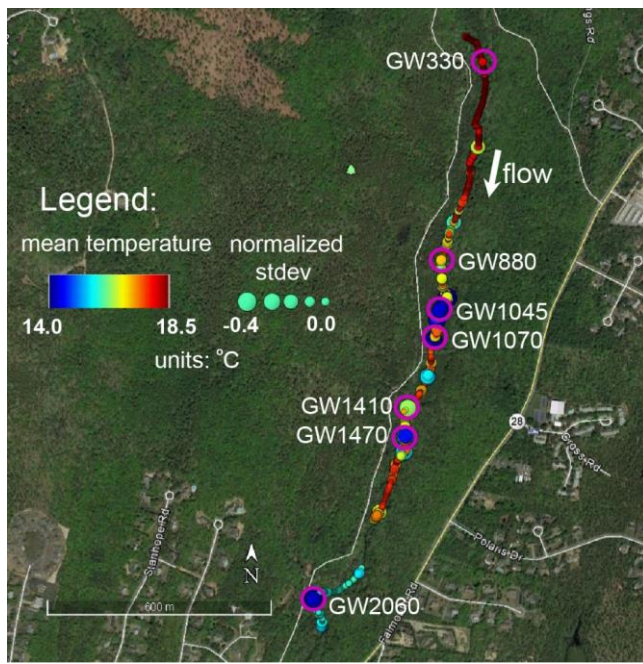


Figure 4 in Rosenberry et al. (2016), panel b) shows summary fiber-optic distributed streambed interface temperature data were collected at 1-m scale along the streambed interface. Larger dots over time to indicate reduced thermal variance as buffered by preferential groundwater discharge, while points of zones using reduced thermal variance and relatively cold mean temperatures also suggest strong groundwater influence. Discharge zones explored in detail are noted numerically following the nomenclature of Rosenberry et al. temperature metrics. Apparent streambed discharge zones with a range of temperature characteristics were directly sampled for dissolved oxygen, specific conductivity, and stable water isotopes, and labeled as downstream distance from a common upstream point (e.g. GWxxxx from the fish ladder). This Figure is modified from Rosenberry et al. (2016).

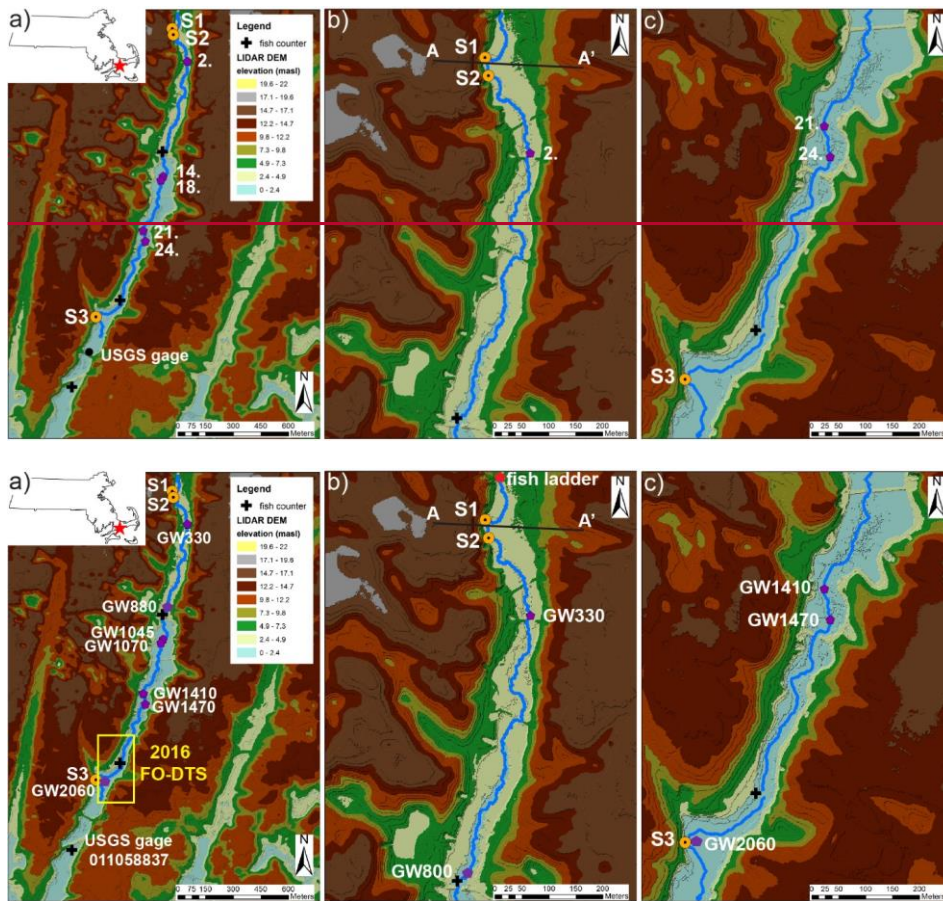


Figure 2. Lidar elevation data show the linear valley terrain of: a) The lower Quashnet River reach with Spawn (S1, S2, S3) locations (orange circles) and major open valley seepage zones (purple circles) identified. All seepage zones are labeled as downstream distance from the stream crossing/fish ladder located at the upper extent of this image. Panel b) shows the tighter upper valley zone where Spawn 1 and 2 are located at the base of a steep cut bank and the topographic transect of Figure 1 is noted. Panel c) displays the lower open valley reach where Spawn 3 is located along a cut bank.



Figure 3. Images collected in February 2016 a) the cut bank alcove at Spawn 1, b) open valley seepage [Location 14/15GW1045](#), and c) fresh cut bank slumping and visible seepage Spawn 3. Panel d) is an image from the underwater video collected in fall 2015 of spawning trout in the [Spawn 1](#) alcove pictured in panel a), showing several fish clustered around the sandy zone directly at the base of the cut bank.

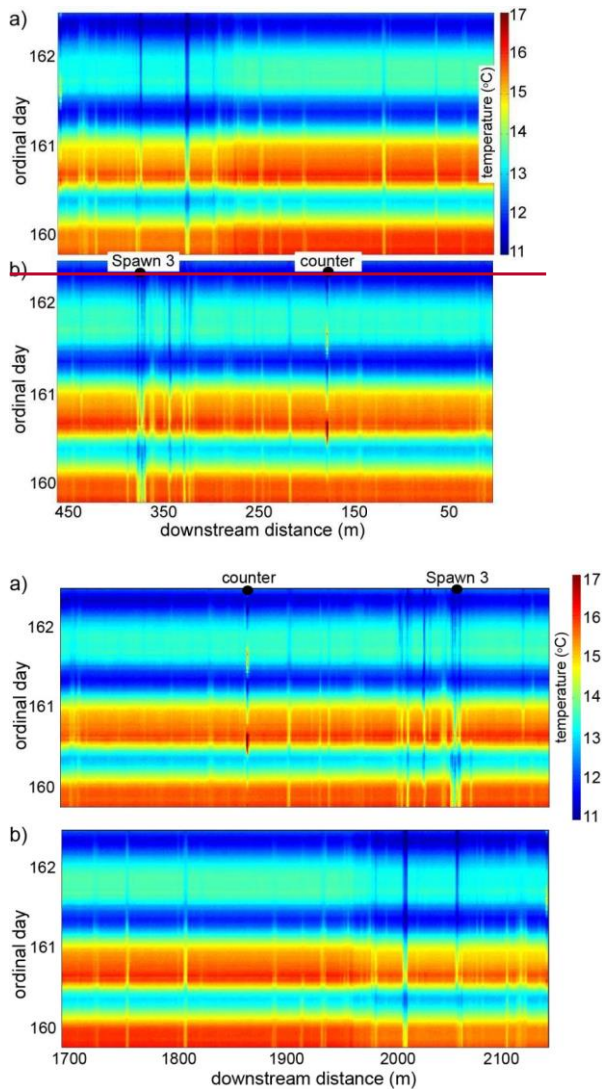
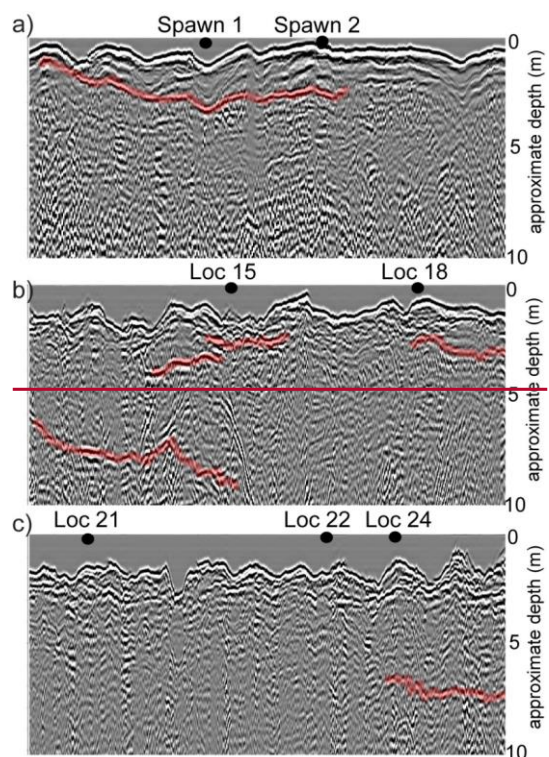
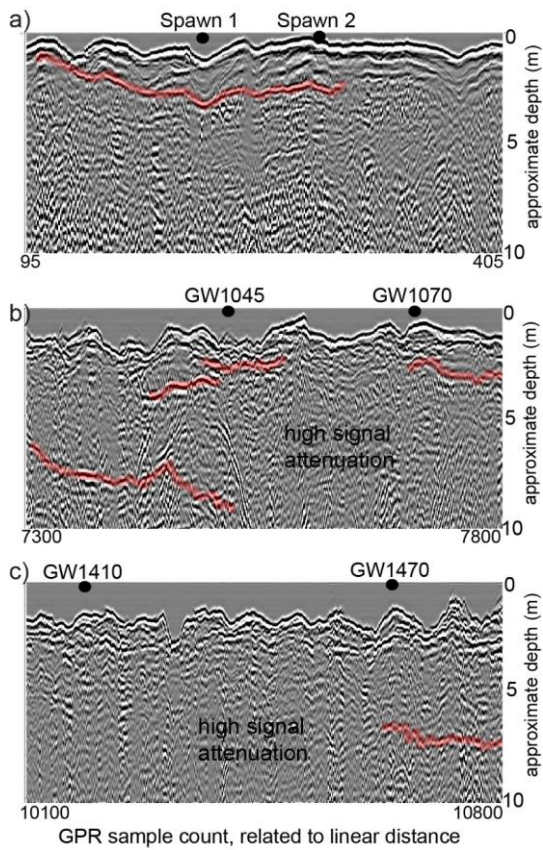


Figure 4. Fiber-optic-distributed temperature data collected starting at an arbitrary stream point from approximate channel distance 1700 to 2160 m along a) the downstream left bank and b) the downstream right bank through the Spawn 3 meander bend area (see Figure 2c for location), and b) the downstream left bank. The persistent vertical bands of cooler (blue) colors/relatively cool temperatures indicate discrete groundwater discharge; some. Some larger zones display a thermal signature on both bank bank cables, while smaller discharges may be bank-specific.

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GPR sample count, related to linear distance

Figure 5. Quashnet River thalweg ground penetrating radar profiles were collected in the vicinity of: a) Spawn 1 and 2; b) open valley seepage ~~Locations 15~~GW1045 and ~~18~~GW1070; and c) open valley seepage ~~Locations 21, 22~~GW1410 and ~~24~~GW1470. Stronger apparent reflectors are highlighted in red, and likely indicate sediment layer boundaries (e.g. sand/gravel and peat).

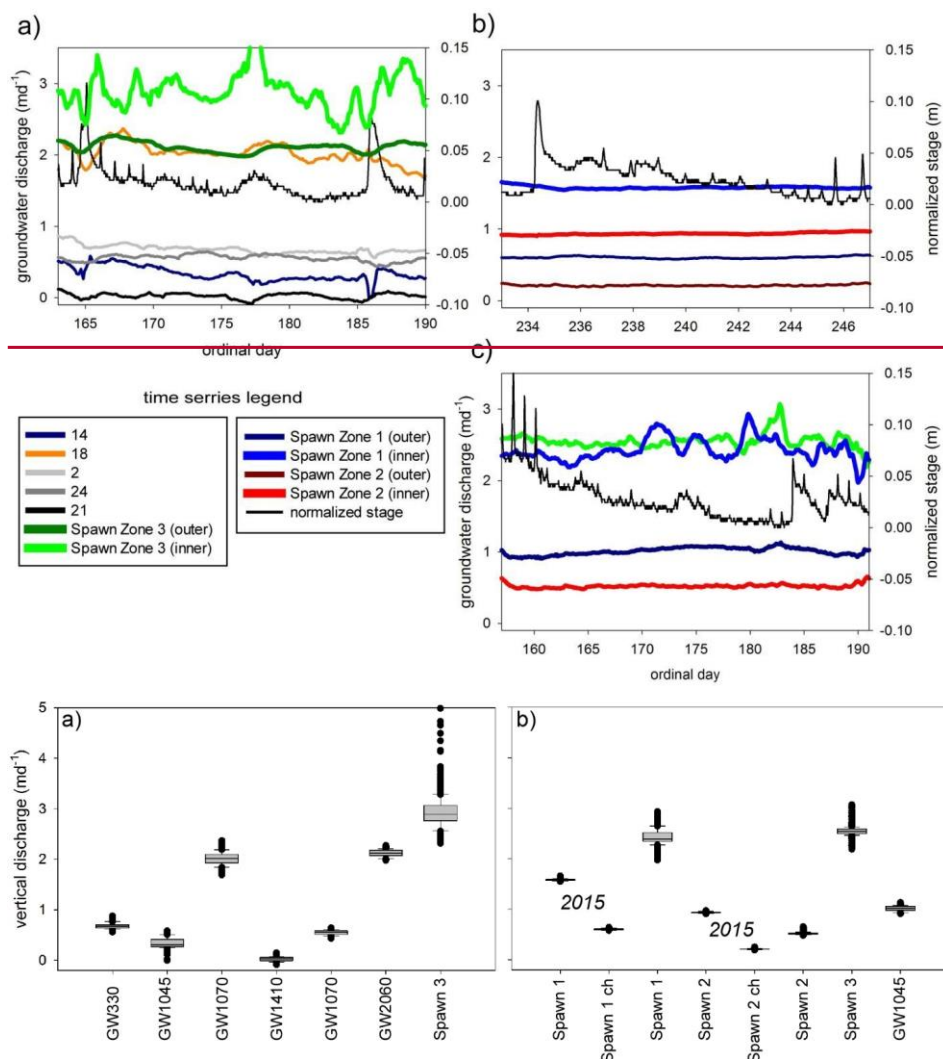
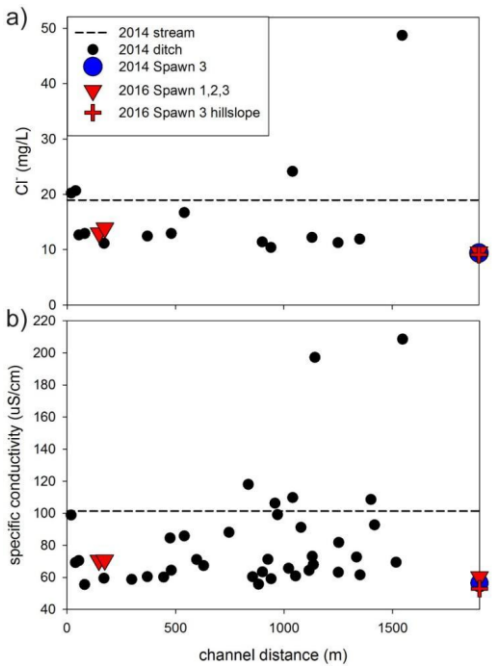


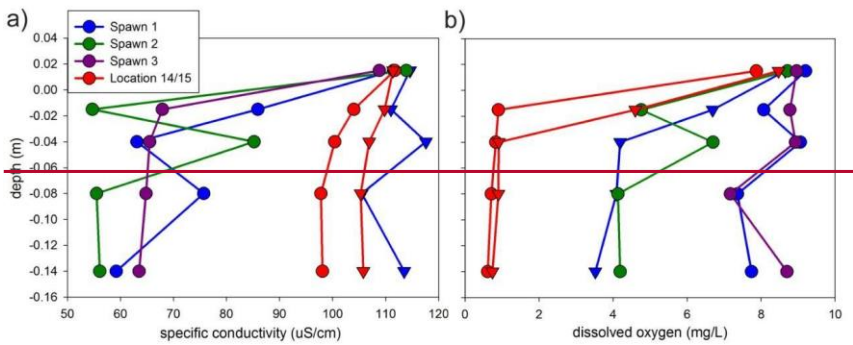
Figure 6. The Quashnet River USGS gaging station stage is compared to Panel a) displays summary box plots of sub-daily time series of (June 11 to July 13, 2014) vertical groundwater flux discharge rate estimates for the open valley discharge and Spawn 3 bank locations. Panel b) shows vertical discharge rates collected against the banks and toward the channel at various strong seepage Spawn zones in a) June 2014, b) 1 and 2 (August 21 to September 13, 2015, and c) and from locations against the bank in all three Spawn locations and GW1045 (June 5 to July 9, 2016).

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Figure 7. Drainage ditch chemistry throughout the lower Quashnet showing a) Cl^- , and b) specific conductance, collected in June 2014 just above the confluence with the main channel. Data are plotted as distance from the upper flood control structure in the narrow valley reach and compared to groundwater seepage data collected in preferential spawning locations and a hillslope piezometer.



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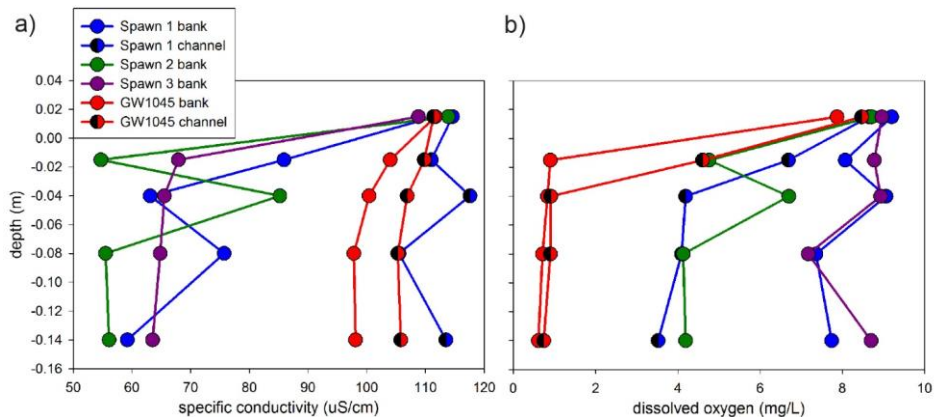


Figure 8. Minipoint pore-water chemistry data showing high spatial resolution profiles of a) specific conductance, and b) dissolved oxygen, collected in June 2016 at the major seepage alcoves. Triangle symbols indicate data collected farther toward the thalweg from the respective alcove bank, and all profiles include a local stream water sample taken just above the streambed interface.

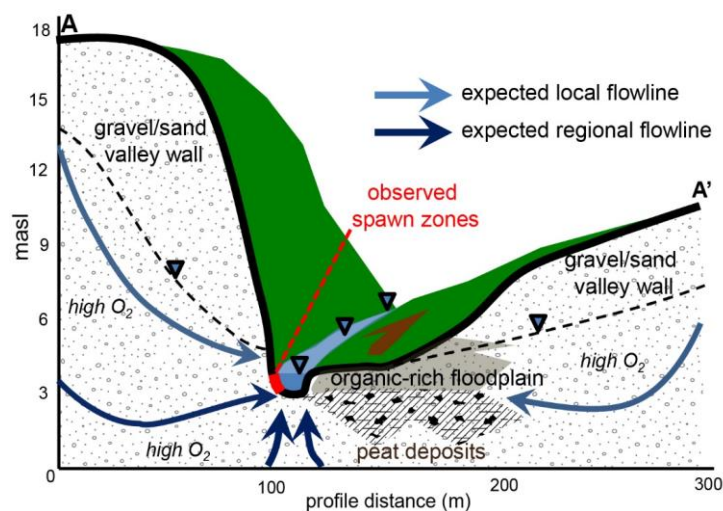


Figure 9. Based on previously published USGS work (e.g. Modica, 1999; Winter et al., 1998), the conceptual model in panel a) displays how groundwater discharge to lowland streams is

1556 expected to include locally sourced lateral groundwater discharge through valley wall features
1557 and more regionally sourced groundwater discharge vertically through the streambed. The
1558 topographic profile shown here (A-A') is derived directly from lidar data in the vicinity of
1559 observed preferential brook trout spawning habitat shown in Figure 2. In contrast to the sand and
1560 gravel valley walls, multiple methodologies used for this study indicate wider valley zone
1561 sediments to be rich in organic material, including buried peat deposits, consistent with known
1562 regional geology.

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1564 **Supplemental**

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1566 Supplemental Video S1. Underwater video of brook trout spawning in the fall of 2015 (still
1567 image Figure 3d).