Review #1 Author Response

Please see our specific revisions below listed in black text:

The authors have addressed my major concerns. This is an excellent, comprehensive study, and the authors should be commended on assembling all the components into a manuscript that will be appreciated by a wide, interdisciplinary audience. I have no further recommendations for major changes. However, I have identified many stylistic issues and minor problems that need to be rectified in the text and especially in the figures. These recommendations and suggestions are listed in the specific comments below.

Thank you for the positive assessment. We have addressed specific suggestions as detailed below.

Specific comments

Line 68: I recommend not capitalizing "Unmanned Aircraft Systems". Check for other examples throughout the manuscript. done

Line 80: Check spelling of "Mathews". Corrected

Line 81: No need to capitalize "Northeastern". fixed

Line 83: Use "cold-water" as a modifier of "habitat". done

Line 120: Citations should be in chronological order. We now believe the references are formatted per HESS guidelines, throughout

Line 152: "PIT" needs to be defined at first mention. done

Line 226: The acronym "PIT" without the definition can be used here because it will have been

30 described earlier in the manuscript. done

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32 Line 307: Check "pore water were also collected" for subject and verb agreement. This sentence
33 was modified

34

35 Line 321: Specify the wavelength for the thermal IR camera. We added "thermal" infrared to
36 indicate the broader IR spectrum and the model number is listed

37

38 Line 326: Define "SpC" at first mention. done

39

40 Line 331: No hyphen is needed in "locally-recharged" (https://www.merriam-
41 webster.com/words-at-play/6-common-hypercorrections-and-how-to-avoid-them/hyphenating-
42 ly-adverbs). Check entire document for other examples of this kind of incorrect hyphen use (e.g.,
43 "ecologically-based", etc.). changes made

44

45 Line 356: The second sentence in paragraph is a typographical error. I suggest that the first
46 sentence be moved to the beginning of the following section titled "Observations regarding
47 repeat spawning locations". This sentence was deleted. We need to preserve an intro sentence or
48 two for each main section per USGS style, as not to have "hanging titles"

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50 Line 379: Check "data was". It should be "data were". changed

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52 Results: The authors are consistent in reporting their results in the present tense, but this is
53 awkward to me because the conditions and observations are in the past. We have chosen to use
54 the present tense here to be consistent with other recent publications from the group. It seems
55 this falls in the category of "authors choice"

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57 Line 477: "Global Positioning System" does not need to be capitalized. Agreed, changed

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59   Line 598: "over 10 yr+" could be written as "> 10 yr". this was changed

60

61   Table captions: It is awkward to begin these captions with a number. Also, I found these captions
62   (and those of the figures) to be written in a nonstandard manner that makes it difficult to quickly
63   determine the contents of the table that follows. Other manuscripts by Briggs et al. don't have
64   this problem, so the lead author can certainly make the necessary changes. The captions were
65   adjusted as to not start with dates

66

67   Figure 1: Number in scale bar is too small. No need to write "Legend". Can you use arrows to
68   point to the locations of your sites instead of using purple circles, which obscure the temperature
69   dots? The way the captions is written, it is too difficult to quickly determine what the sites
70   labeled with "GW" mean. Honestly, the caption does an inadequate job of describing what this
71   figure depicts. For example, the first sentence of the caption is a statement of the methods. See
72   other papers written by Briggs et al. for better examples of how captions should be written.
73   These temperature data were from DTS, correct? This needs to be stated in the caption. These
74   data are plotted in Google Earth, where it is not obvious how to change the scale bar size (this
75   would be easy in Arcmap. The scalebar has been now enhanced in post processing. The purple
76   circles have been replaced with arrows and the caption adjusted to be more clear and completer.

77

78   Figure 2: Need "MA" on inset map. The numbers in the scale bars are too small. Just draw a
79   simple black bar in a white box to indicate 200 m. This will need to be done in each panel
80   because the scales are different. No need to write "Legend"; make the text larger in the contents.
81   It's still too hard to read. Can you define the purple dots and orange circles in the legend? The
82   transect A-A prime is not mentioned in the caption. Remove it if it is unimportant. It would be
83   helpful to have rectangles on Figure 2a to show the extents of the insets (Figures 2b and 2c). The
84   color scheme is problematic because there is no natural progression of colors. This may seem
85   like a minor point, but it does help the reader when interpreting the range from high to low.
86   Finally, the writing of the caption is awkward and not standard with "Panel c)" written in a
87   grammatically incorrect style. Please other papers by Briggs et al. for correct examples of how
88   captions are written.

89   We have made these changes where practical. Scale bars were replaced as requested. Font size is
90   practically limited by the size of the Figure, but it has been enhanced throughout the legend. The
91   groundwater discharge symbols and spawn locations have been added to the legend, and
92   "legend" deleted.The transect is important to Figure 9, as described now in legend. The inset
93   boxes were attempted, but make an already busy Figure too cluttered. Color schemes are in some
94   part a personal preference of the reader. We experimented with several gradational schemes for

95   this, but feel the current scheme allows the floodplain to most clearly be distinguished from the
96   valley wall, which is directly relevant to the main theme of the paper. The key for color vs
97   elevation is clearly labeled.

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99   Figure 3: See other comments about how captions should be written. Other papers by Briggs et
100   al. have excellent examples of nicely written captions. The caption has been edited

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102   Figure 4: This caption is written better, but it will help to use () around the panel letters. Note
103   that a single parenthesis should be used where there is no text to the left (i.e., in a numerical list):
104   () added

105

106   1)

107   2)

108   3)

109

110   or

111

112   a)

113   b)

114   c)

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116   Figure 5: Please write captions in a standard manner as is appropriate for a scientific journal. The
117   first line of the caption is not a methodological statement; it should be a concise statement
118   describing the contents of the figure. Also, it would be very helpful to provide a more explicit
119   description of the distance on the x-axis, even if it is approximate. See other comments about
120   how parentheses should be used if they are enclosing parenthetical information. Caption edited.
121   We do not feel comfortable adding an approximate x-axis due to variable float speed of the
122   radar, but the locations of interest (spawn zones, discharge zones) were directly marked in the
123   record and explicitly shown here.

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125 Figure 6: See comments for other figures on how captions should be written. Caption edited

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127 Figure 7: This caption is better than the others, but the panel labels need be enclosed in
128 parentheses in the caption. OK

129

130 Figure 8: The caption is poorly written. For example, to begin a caption with "Based on..." not
131 appropriate. There is no reason to provide citations in this caption. Perhaps the authors
132 mistakenly used a rough draft for all of these captions because Briggs et al. in their other papers
133 do a fine job of writing captions. Also, what is the brown arrow in the green area? Can you
134 remove this brown arrow? This figure also needs an arrow showing the direction of stream flow.
135 It's a difficult figure to understand because it's a 2-D graph with a 3-D figure inserted within it. A
136 flow direction arrow over the stream would help. We assume you refer to Figure 9 here, and the
137 suggestions have been adopted

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139 **Review #2 Author Response**

Please see our specific revisions below listed in black text:

The revised paper has been significantly improved from the previous version. There are only a
few minor edits that are needed:
Line 236: This sentence is hard to understand, please consider rephrasing. Yes we were missing
a word in this sentence, and that has been fixed.
Line 340-341: please use the correct format for dates. Dates were changed to European format
throughout
Line 358: this is not a complete sentence and it appears to be a duplicate section title. Yes this
was a duplicate title, and has been deleted
Line 530: change "course" to "coarse" change made

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142 **Hydrogeochemical Controls on Brook Trout Spawning Habitat in a Coastal Stream**
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144

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176   **Abstract:**
177   Brook trout (*Salvelinus fontinalis*) spawn in fall, and overwintering egg development

178   can benefit from stable, relatively warm temperatures in groundwater seepage zones. However,

179   eggs also are sensitive to dissolved oxygen concentration, which may be reduced in discharging

180   groundwater (i.e. seepage). We investigated a 2-km reach of the coastal Quashnet River, Cape

181   Cod, Massachusetts, USA, to relate preferred fish spawning habitat to geology, geomorphology,

182   and discharging groundwater geochemistry. Thermal reconnaissance methods were used to

183   locate zones of rapid groundwater discharge, which were predominantly found along the center

184   channel of a wider stream valley section. Pore-water chemistry and temporal vertical

185   groundwater flux were measured at a subset of these zones during field campaigns over several

186   seasons. Seepage zones in open valley sub-reaches generally showed suboxic conditions and

187   higher dissolved solutes compared to the underlying glacial outwash aquifer. These discharge

188   zones were cross-referenced with preferred brook trout redds, evaluated during 10 yr of

189   observation, all of which were associated with discrete alcove features in steep cut banks where

190   stream meander bends intersect the glacial valley walls. Seepage in these repeat spawning zones

191   was generally stronger and more variable than open valley sites, with higher dissolved oxygen

192   and reduced solute concentrations. The combined evidence indicates that regional groundwater

193   discharge along the broader valley bottom is predominantly suboxic due to the influence of near-

194   stream organic deposits; trout show no obvious preference for these zones when spawning.

195   However, the meander bends that cut into sandy deposits near the valley walls generate strong,

196   oxic seepage zones that are utilized routinely for redd construction and the overwintering of trout

197   eggs. Stable water isotopic data support the conclusion that repeat spawning zones located

198   directly on preferential discharges of more localized groundwater. In similar coastal systems

199   with extensive valley peat deposits, specific use of groundwater discharge points by brook trout

200   may be limited to morphologies such as cut banks where groundwater flowpaths do not

201   encounter substantial buried organic material and remain oxygen rich.

Introduction

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 handheld cameras, piloted aircraft, and the rapidly evolving capabilities of unmanned aerial systems~~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 coldwater fish habitat such as summer thermal refugia (Dugdale et al., 2015). However, surface thermal surveys alone do not indicate groundwater flowpath dynamics or the suitability of interface aquatic habitat (Briggs et al., 2018a).

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, coldwater 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 (Matthews and Berg, 1997). The use of groundwater upwelling zones as thermal refugia is further complicated by competition with aggressive invasive species (to the ~~Northeastern~~ northeastern USA) such as brown trout that compete with native trout for resources (Hitt et al., 2017). Streams at higher elevations may support the persistence of reach-scale cold water habitat

225 where point-scale thermal refugia are not needed under current climatic conditions, serving as

226 vital "climate refugia" against rising air temperatures (Isaak et al., 2015). In systems with

227 reliably cold channel water in summer, which can also exist at low elevations when heavily

228 influenced by discharging groundwater, salmonid fish may directly use groundwater seepage

229 zones for spawning rather than thermal refuge.

230   Brook trout (*Salvelinus fontinalis*) are a species of char that are native to eastern North

231 America, from Georgia to Quebec (MacCrimmon and Campbell, 1969). Populations have been

232 stressed by warming temperatures and reduced water quality, particularly in low-elevation areas

233 (Hudy et al., 2008). Stream network-scale tracking of fish has indicated brook trout directly

234 utilize stream confluence mixing zones and preferential groundwater discharge to survive warm

235 summer periods (Baird and Krueger, 2003; Petty et al., 2012; Snook et al., 2016). Additionally,

236 brook trout spawn in the fall, and eggs deposited in redds develop over the winter before

237 hatching in spring (Cunjak and Power, 1986). Oxygen use by the shallow buried embryos

238 increases over the period of development (Crisp, 1981), and therefore DO concentration is a

239 critical parameter of the pore waters in which the eggs are bathed. Several studies have

240 demonstrated the importance of hyporheic downwelling in increasing shallow oxygen

241 concentrations, including at salmonid redds, where deeper streambed pore water is generally

242 reduced in DO (e.g. Buffington and Tonina 2009; Cardenas et al. 2016; Harvey et al.,

243 2013). Several studies have demonstrated the importance of hyporheic downwelling in

244 increasing shallow oxygen concentrations specifically at salmonid redds when streambed pore

245 water is generally reduced in DO (e.g. Buffington and Tonina 2009; Cardenas et al. 2016). Fine

246 sediments can reduce the efficacy of hyporheic DO exchange in spawn zones (Obruca and

247 Hauer, 2016), and are actively cleared by trout during the spawning process ((Montgomery et al.,

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248 1996).

249   The importance of hyporheic exchange to salmonid spawning may be limited in the

250 lowland streams that are expected to harbor native cold-water species in the 21st century: those

251 with strong groundwater influence. Groundwater upwelling reduces the penetration of hyporheic

252 flow from surface water (Cardenas and Wilson, 2006) and may shut down hyporheic flushing in

253 redds (Cardenas et al., 2016). Where hyporheic exchange does introduce oxygenated channel

254 water into the shallow streambed, the downward advection of heat associated with near-freezing

255 surface water in winter will also cool streambed sediments (Geist et al., 2002), potentially

256 impairing egg development. Coaster brook trout, a life-history variant of native brook trout

257 exhibiting potadromous migrations within the Great Lakes, have been shown to specifically

258 prefer groundwater discharge zones for building redds (Grinsven et al., 2012). The development

259 of trout in winter has been found to be positively correlated with warmer stream water

260 temperatures as influenced by groundwater seepage (French et al., 2016), and therefore spatially

261 discrete groundwater discharge zones with adequate DO may form preferred brook trout

262 spawning habitat (Curry et al., 1995).

263   Multiscale physical and biogeochemical factors influence temperature and DO

264 concentrations along groundwater flowpaths. In river valleys, discharge to surface water of

265 locally recharged groundwater is expected to emanate from more shallow, lateral flowpaths

266 controlled by local topography (Modica, 1999; Winter et al., 1998). Shallow groundwater

267 flowpaths, particularly those within approximately 5 m of the land surface, will be more sensitive

268 to annual air temperature patterns and longer term warming trends due to strong vertical

269 conductive heat exchanges (Kurylyk et al., 2015b). The distance of seeps from upgradient

270 groundwater recharge zones will also affect seepage temperature dynamics and associated

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271 aquatic ecosystems due to future changes in surface and recharge temperature (Burns et al.,

272 2017). Therefore, working backwards from thermal anomalies into the landscape is critical to

273 understanding the thermal stability of current and future point-scale preferential brook trout

274 habitat (Briggs et al., 2018b). The complimentary methodology of geophysical remote sensing,

275 geochemical sampling, and vertical bed temperature time series can indicate the physical and

276 chemical properties of groundwater flowpaths that source preferential ~~disharge~~discharge zones

277 utilized routinely by fish for spawning.

278   Coarse-grained mineral-dominated aquifers with little fine particulate organic matter and

279 low dissolved organic carbon supply tend to result in generally oxic groundwater conditions

280 (Back et al., 1993). The sandy surficial aquifer of Cape Cod, where our investigation took place,

281 is a classic example of a mineral soil-dominated flow system (Frimpter and Gay, 1979). Flow of

282 groundwater through near-stream organic deposits, however, can result in inverted redox

283 gradients toward the upwelling interface, such that groundwater discharged to surface water is

284 reduced in DO (Seitzinger et al., 2006). In sandy glacial terrain with superimposed peatland

285 deposits, the specific flow patterns of groundwater to surface water in relation to buried peat will

286 influence groundwater discharge biogeochemistry. Krause et al. (2013) found that streambed

287 groundwater seepage was strongly reduced in DO in zones with peat deposits, likely due to an

288 increase in both near-stream residence time and localized source of dissolved organic carbon.

289   Interdisciplinary collaborations between physical and biological scientists are useful to

290 better understand how cold-water species utilize groundwater discharge-influenced stream

291 habitat, and the larger landscape-scale controls on discharge characteristics. While previous

292 hydrogeological research in the coastal stream used for this study had focused on locating and

293 quantifying discrete groundwater discharge (e.g. "cold anomalies", Hare et al., 2015; Rosenberry

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294 et al., 2016), here we endeavor to understand the hydraulic and biogeochemical controls on

295 seepage zone distribution utilized directly by native brook trout. In this groundwater-dominated

296 stream (e.g. likely climate refugia), brook trout do not need to occupy discrete inflows for

297 summer thermal refugia, but do favor certain upwelling zones for fall spawning. We compare

298 over a decade of visual survey and electronic fish passive integrated transponder (PIT)-tag

299 dropout data regarding repeat brook trout spawning locations to a comprehensive physical and

300 chemical characterization of groundwater seepage zones across 2-km of stream to:

301   1. Identify repeat brook trout spawning locations, and determine if they are directly

302   associated with the preferential discharge of groundwater through interface sediments.

303   2. Develop a hydrogeochemical characterization of trout-preferred groundwater discharge

304   zones that can aid in their identification in other less-studied systems and potential

305   inclusion in stream habitat restoration efforts.

306 **Site Description and Previous Hydrogeologic Characterization**

307   Cape Cod is a peninsula in southeastern coastal Massachusetts, USA, composed

308 primarily of highly permeable unconsolidated glacial moraine and outwash deposits. The largest

309 of the Cape Cod sole-source aquifers occupies a western (landward) section of the peninsula

310 (LeBlanc et al., 1986), and is incised by several linear valleys that drain groundwater south to the

311 Atlantic Ocean via baseflow-dominated streams (Figure 2a). Strong groundwater discharge to

312 one such stream, the Quashnet River, supports a relatively stable flow regime that has averaged

313 0.49 +/- 0.15 (SD) $m^3 s^{-1}$ from 1986-2015 (Rosenberry et al., 2016). The lower Quashnet River

314 emerges from a narrow sand and gravel valley to a broader area with well-defined lateral

315 floodplains. Historical cranberry farming practices, abandoned in the 1950s, have modified the

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316   stream corridor (Barlow and Hess, 1993). Primary modifications included straightening of the

317   main channel (reducing natural sinuosity), installation of flood-control structures, incision of

318   shallow groundwater drainage ditches in the lateral peatland floodplain, and widespread

319   application of sand to the floodplain surface. The current bank-full width of the main channel

320   averages approximately 4 m.

321   The Quashnet River has long been recognized as critical habitat for a naturally reproducing

322   population of native sea-run brook trout (Mullan, 1958) with a genetically distinct population

323   (Annett et al., 2012). Efforts to restore trout habitat by the group *Trout Unlimited* and others

324   have been ongoing for over 40 yr (Barlow and Hess, 1993). These efforts include the removal of

325   flood-control structures and planting of trees along the main channel, and addition of wood

326   structures to stabilize banks and provide cover from airborne predators. Further, the

327   Commonwealth of Massachusetts purchased 12.5 hectares in 1956 and an additional 146

328   hectares along the lower Quashnet River in 1987 and 1988 to protect the area from development.

329   The Massachusetts Division of Fisheries and Wildlife has been monitoring trout populations

330   since 1988 and movement since 2007.

331   Groundwater influence on stream temperature is pronounced, particularly over the 2-km

332   reach above the USGS gage (Briggs et al., 2018a), below which stream stage is tidally affected.

333   Ambient regional groundwater temperature is approximately 11 $^{\circ}$C (Briggs et al., 2014), and

334   strong conductive and advective exchange with the proximal aquifer maintains surface water

335   temperature well below the lethal threshold for brook trout (maximum weekly average

336   temperature >23.3 °C, Wehrly *et al.* 2007). Therefore point-scale thermal refugia are not a

337   current concern in this system, as the stream supports system-scale cold-water habitat that is

338   likely to persist into the future and serve as warming "climate refugia" (Briggs et al., 2018a). In

339 winter, seepage zones can be located as relatively warm anomalies increasing and buffering

340 surface water temperatures from ambient atmospheric influence.

341   Previous work has measured relatively large net gains in streamflow over the lower Quashnet

342 River (Barlow and Hess, 1993; Rosenberry et al., 2016), attributed to groundwater discharge

343 through direct streambed seepage and harvesting of groundwater from the floodplain platform

344 via relic agricultural drainage ditches. Deployments of fiber-optic temperature sensing (FO-DTS)

345 cables along the thalweg streambed interface indicate the greatest density of focused seepage

346 zones occurs along the broader valley area approximately 1 km upstream of the USGS gage

347 number 011058837; this zone coincides with the largest gains in net streamflow (Hare et al.,

348 2015). Based on the streambed interface temperature data presented by Rosenberry et al. (2016),

349 Figure 1 shows how temperature-sensitive fiber optic cables have been used to pinpoint possible

350 groundwater discharge zones based on anomalously cold mean temperature and/or reduced

351 thermal variance. Focused evaluation of FO-DTS anomalies with physical seepage meters and

352 vertical temperature profilers confirmed localized, meter-scale seepage zonation along the

353 streambed where discrete colder zones indicated through heat tracing showed approximately 5

354 times the groundwater discharge rate of adjacent sandy bed locations only meters away

355 (Rosenberry et al., 2016). Active heating of wrapped FO-DTS cables deployed vertically within

356 an open valley streambed seepage zone indicated true vertical flow to at least 0.6 m into the bed

357 sediments (Briggs et al., 2016), an expected characteristic of more regional groundwater

358 discharge (Winter et al., 1998), rather than that driven by valley topography local to the river.

359 Hyporheic exchange in the lower Quashnet River system is superimposed on the general upward

360 hydraulic gradient to the stream, and therefore reduced to a thin, shallow hyporheic exchange

361 zone (e.g. < 0.1 m depth) along the thalweg by these competing pressures (Briggs et al., 2014),

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362   as has been simulated for similar stream systems (e.g. Cardenas and Wilson 2006).

363   **Methods**

364   A combination of fish tagging and visual spawning observations, heat tracing,

365   geophysical surveys, and focused pore-water sampling was used to investigate the interplay

366   between the locations of preferential brook trout spawning and the local hydrogeology. For

367   consistency between varied methods and years of data collection, all sample locations are

368   spatially referenced as downstream channel distance from the fish ladder river crossing at the

369   upper end of the study reach (Figure 2).

370   *Observations regarding repeat spawning locations*

371   Observations regarding discrete repeat brook trout spawning locations were made

372   opportunistically as part of an ongoing PIT (Passive Integrated Transponder) tagging study of the

373   native reproducing population of the Quashnet River. Large-scale trout movements are

374   continuously monitored in the lower Quashnet River at 3 stationary fish counting sites (Figure

375   2a). However the spatial resolution of these counting sites, separated by hundreds of meters, is

376   not adequate to study how brook trout utilize specific decimeter- to meter-scale groundwater

377   discharge zones. For this finer scale characterization, dropped fish tags have also been located

378   through roving surveys using a handheld portable PIT antenna (Biomark, Inc.) conducted in

379   spring and fall since 2007. The dropout of PIT tags from the fish body is a process that is more

380   likely to happen during spawning behavior in salmonids (Meyer et al 2011), so dropped tags

381   were located electronically and spatially mapped to reveal discrete zones of repeat spawning.

382   Although these roving surveys do not yield the temporal continuity of the instream counting

383   gates, clustering of dropped tags can be mapped at the sub-meter scale, presumably directly at

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384   trout redds. In addition, spawning brook trout were located visually during annual fall data

385   collection events by Massachusetts Fish and Wildlife Staff, with redd development behavior

386   captured within one seepage feature by underwater video in 2015 using a GoPro Hero camera

387   (San Mateo, CA). We refer to the 3 most prominent sites of brook trout spawning within the

388   study reach as Spawn 1 (113 m), Spawn 2 (146 m), and Spawn 3 (2062 m), from upstream to

389   downstream, respectively (Figure 2).

390   *Spatial mapping of preferential groundwater discharges*

391   To augment existing streambed interface thermal surveys for preferential groundwater

392   discharge (e.g. Rosenberry et al., 2016; Figure 1) and the bank-dependence of discharge location,

393   ruggedized fiber-optic cables suitable for stream use were deployed in the river along the base of

394   each bank from 1700 to 2160 m on ~~June~~ 10 June through ~~June~~ 12 June ~~.~~ 2016 (Figure 2a). Two

395   separate cables weighted with stainless steel armoring were installed directly along the foot of

396   each bank on top of the streambed interface. Single-ended measurements made at the 1.01 m

397   linear spatial sampling scale were integrated over 5-min intervals on each channel by an Oryx

398   FO-DTS control unit (Sensornet Ltd.). During the same period, data were also collected along a

399   high-resolution wrapped fiber-optic array for a dataset described in Kurylyk et al. (2017) but not

400   shown here; this experimental setup resulted in measurements for each channel of 4 instrument

401   channels recorded at 20-min intervals. Calibration for dynamic instrument drift was performed

402   automatically using an approximately 30-m length of cable for each channel submerged in a

403   continuously mixed ice-bath and monitored with an independent Oryx T-100 thermistor.

404   *Quantification of vertical groundwater discharge rates*

405   Once preferential discharge locations are located along the streambed with FO-DTS,

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406 actual vertical discharge rates can be assessed with a variety of methodologies (Kalbus et al.,

407 2006). Temporal patterns in groundwater discharge flux rate can indicate source flowpath

408 hydrodynamics, and be derived from bed temperature time series using vertical temperature

409 signal transport characteristics, as reviewed by Rau et al., (2013). Custom "1DTempProfilers"

410 designed specifically for the quantification of groundwater discharge (Briggs et al., 2014) were

411 used to monitor streambed temperature over time along a shallow vertical profile. Profilers were

412 deployed within a subset of the thermal anomalies previously identified with FO-DTS. The

413 profiler deployment locations were chosen to represent a range of preferential groundwater

414 discharge rates/characteristics based on the on the observed FO-DTS temperature anomalies, e.g.

415 anomalies of varied mean temperature and buffering effect (Figure 1). These preferential

416 groundwater discharges were located: 330, 880, 1045, 1070, 1410, 1470, and 2060 m

417 approximate downstream distance from the fish ladder crossing. These groundwater discharge

418 locations are referred to with the prefix "GW" followed by the meter mark for the remainder of

419 the manuscript, such that major streambed seep 330 m downstream of the fish ladder is referred

420 to as "GW330". Data were collected at various locations from ~~June~~ 11 June to ~~July~~ 13 ~~in~~ July

421 2014; ~~August~~ 21 August to ~~September~~ 13 September ~~in~~ 2015; and ~~June~~ 5 June to ~~July~~ 9 July ~~in~~

422 2016. These deployments included installation of 1DTempProfilers at the nearbank and channel

423 sides of observed repeat spawning zones.

424   Individual thermal data loggers (iButton Thermochron DS1922L, Maxim Integrated)

425 were waterproofed with silicone caulking and inserted horizontally into short slotted-steel pipes

426 (0.025 m diameter). The shallow thermal profilers were driven vertically into the streambed so

427 that sensors were positioned at some combination of 0.01, 0.04, 0.07, and 0.11 m depths. Data

428 were collected at temporal intervals of 0.5 hr in 2014, 2015, and 1 hr in 2016. Rosenberry et al.

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429 (2016a) found that when a subset of the 2014 streambed temperature data presented here were

430 analyzed using the diurnal signal amplitude attenuation models employed by VFLUX2 (Irvine et

431 al., 2015), a near 1:1 relation was found in comparison to physical seepage meter measurements

432 of groundwater discharge ranging from 0.5 to 3 md^{-1}. A similar diurnal signal-based in-situ

433 streambed thermal parameter estimation is used here.

434 *Streambed groundwater discharge and spawning zone pore water characterization*

435   Subsurface water samples were collected for chemical analysis at 7 major open valley

436 seepage locations and 3 repeat spawn locations. Geochemical data collection occurred in 2014

437 and 2016 along with the 1DTempProfiler deployments, while stable water isotope data were

438 collection in August 2017. For geochemical sampling, 0.0095 m (nominal) stainless steel

439 drivepoints were inserted to depths of 0.3, 0.6, and/or 0.9 m and Masterflex Norprene tubing was

440 attached to the drivepoint. A peristaltic pump was used to extract pore water samples until free of

441 obvious turbidity (typically requiring 3 min of pumping) after which the pumping rate was

442 slowed and, the groundwater samples were collected by pumping into 60-mL HDPE syringe

443 barrels. First an unfiltered sample for specific conductivity was pushed from the syringe into a

444 30-mL HDPE Nalgene sample bottle. Second, a filtered sample for anion analysis was collected

445 after attaching a 0.2-µm pore size (25-mm diameter) Pall polyethersulfone filter to the syringe.

446 Lastly, the pumping rate was slowed again and an overflow cup was attached to the norprene

447 sample tubing and held upright until overflowing, at which point DO was measured by a field

448 colorimetric test using the manufacturer's evacuated reagent vials (Chemetrics V-2000). DO

449 concentrations were read twice and the test repeated using an alternative vial kit if results were

450 near the concentration range limit or out of range. The collected samples were kept cool and out

451 of the light and analyzed for Cl$^-$ upon return to the laboratory using standard ion

18

452   chromatographic techniques.

453   In addition to the drivepoint samples, pore water samples were also collected in June

454   2016 from shallow depths 0.015, 0.04, 0.08 and 0.15 m below the streambed surface at locations

455   GW1045 and Spawn 1, 2, 3 using MINIPOINT samplers (e.g. Harvey and Fuller 1998). Water

456   was pumped simultaneously from all depths using a multi-head pump that withdrew small-

457   volume samples (15 mL) at low flow rates (1.5 mL min^{-1}) to minimize disturbance of natural

458   subsurface fluxes and chemical gradients. Pumped lines terminated at press-on luer fittings that

459   were pushed onto 0.2-µm pore size (25-mm diameter) Pall polyethersulfone filters. Samples for

460   specific conductivity were collected whereas filtered samples were collected for anions in

461   prelabeled 20-mL LDPE plastic scintillation vials with Polyseal^{TM} caps. Sample lines were then

462   attached to overflow cups and dissolved oxygen concentrations were measured as described

463   above.

464   During a follow-up field effort in August, 2017 streambed pore water samples were

465   collected at the Spawn sites and at GW1045, GW1140 (approximately 70 m downstream of

466   GW1070), and GW1470. Additionally, two large hillslope springs were identified along the edge

467   of the riparian zone, upstream of Spawn 1, using a handheld thermal infrared camera (FLIR

468   T640, FLIR Systems, Inc.). These exposed springs were sampled to identify a localized hillslope

469   groundwater signature that would not be impacted by valley-floor peat deposits. Samples were

470   drawn from push-point piezometers installed from 0.2-0.44 m below the sediment interface, with

471   deeper samples collected in the hillslope springs to avoid surface organic material. Pore water

472   was evaluated for specific conductivity (SpC), DO, and stable water isotopes. Isotope samples

473   were analyzed by the U.S. Geological Survey Stable Isotope Laboratory using dual-inlet isotope-

474   ratio mass spectrometry. A substantial fraction of regional Cape Cod shallow groundwater

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475 exchanges with the numerous groundwater flow through lakes as it discharges to the coast

476 (Walter and Masterson, 2002). It is therefore assumed that the regional Cape Cod groundwater

477 isotopic signature is likely to indicate evaporative processes (Leblanc et al., 2008), offering a

478 contrasting signal from locally -recharged hillslope groundwaters (no substantial evaporation).

479 Local deuterium excess of contemporary waters can indicate groundwater that has been

480 influenced by evaporation in lakes, and is therefore in disequilibrium with local meteoric waters.

481 Deuterium excess was determined here as: $d\text{-}xs=\delta^2H - 8* \delta^{18}O$ (Dansgaard, 1964).

482   As mentioned previously, historic cranberry farming practices extensively modified the

483 Quashnet River valley including the incision of drainage ditches into the floodplain. Some

484 ditches extend from the valley wall to the main channel, whereas others are shorter or cut at

485 angles. In addition to characterization of pore water, 34 major drainage ditches (observed

486 flowing water) and a stream thalweg profile were spot checked for specific conductivity on 16

487 June , 16 2014 using the SmarTroll probe (YSI). At a subset of these ditch locations, filtered grab

488 samples were collected and analyzed in the laboratory for Cl^- in a similar manner as for the mini

489 and drivepoint samples described above. In June 2016, the dataset was augmented for 5 ditch

490 confluence locations upstream of Spawn 1.

491 *Visualizing streambed sediment geologic structure*

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

493 water/groundwater exchange studies to characterize underlying peat and sandy deposits (e.g.

494 Lowry et al., 2009; Comas et al., 2011) due to strong expected differences in matrix porosity

495 (water content), which can exceed 70% in peat (Rezanezhad et al., 2016). An upstream to

496 downstream GPR profile was collected on July 7 July 2016 using a MALA HDR GX160

497 shielded antenna (MALA GPR, Sweden) hand-towed down the stream center channel with a

20

498   small inflatable watercraft. The locations of major seep and spawning sites were specifically

499   marked on the digital GPR record during data collection. The GPR data were processed using

500   Reflexw software (Sandmeier, Germany) to convert reflection time to interface depth.

501   **Results**

502   The hydrogeochemical characterization of observed, repeat trout spawning zones and

503   other major streambed groundwater discharge zones are contrasted below. ~~Observations~~

504   ~~regarding repeat spawning locations.~~

505   *Observations regarding repeat spawning locations*

506   Out of the dozens of preferential groundwater discharge zones geolocated along the

507   Quashnet River in this and previous work (e.g. Figure 1), brook trout appear to consistently

508   utilize only three discrete streambed locations for repeat spawning activity. These locations

509   coincide with steep cut banks where the river channel approaches the sand and gravel valley wall

510   (Figure 2b,c). Specifically, trout were found to occupy small "scalloped" alcove bank features

511   (Figure 3a) that may be formed by groundwater sapping and subsequent slumping of sandy bank

512   materials. In winter 2016, fresh slumping and direct seepage from the newly exposed sand wall

513   was observed at Spawn 3 (Figure 3c); a larger slump event had filled approximately 1/3 of the

514   scalloped alcove at Spawn 2 by June 2016. Brook trout were observed clustered along the inner

515   bank area at the Spawn 1 location in fall 2015 (Figure 3d), and this spawning behavior was

516   captured using underwater video (Supplemental Video S1).

517   Dropout PIT tags have been found repeatedly in each of the 3 preferential spawn zones.

518   Seven dropout PIT tags were located in the Spawn 3 zone in March 2017, by far the most

519   dropped tags found in any one location since the tracking program began in 2007. The only other

520   obvious scalloped bank features along the 2 km study reach are located at GW1045 (Figure 3b).

521   Compared to the trout spawning zone alcoves along the valley wall cutbanks (e.g. Figure 3a),

522   this open valley seepage alcove was overgrown with watercress and thick (tens of centimeters)

523   loose deposits of organic material.

524   *Spatial mapping of preferential groundwater discharges*

525   As shown in Figure 1, previously collected FO-DTS data ~~was~~ were used to guide data

526   collection at a subset of representative preferential streambed groundwater discharges.

527   Additionally, paired FO-DTS cables were deployed at the base of both stream banks through a

528   lower reach section in 2016 (Figure 2c), revealing differing thermal anomaly patterns (Figure 4;

529   Briggs et al., 2018c). The cable along the downstream-right bank captures a large, 8-m-long

530   cooler zone at Spawn 3 (Figure 4b), and this seepage signature is spatially reduced but visible

531   along the opposing bank (Figure 4a). Other thermal anomalies observed along one bank show

532   little or no signature along the other. Air temperature dropped noticeably over the final 1.5 d of

533   deployment, and smaller cool anomalies that appeared on warm days are no longer captured by

534   the streambed FO-DTS deployment, but the Spawn 3 signature is still visible along both cables.

535   *Quantification of vertical groundwater discharge rates*

536   Ambient streambed temperature signal data can be used to measure streambed thermal

537   conduction parameters (Luce et al., 2013), which is particularly important when applying heat-

538   based methods to quantify upward vertical fluid flux (Rosenberry et al., 2016), compared to

539   downward fluid flux models that generally show less sensitivity to streambed thermal

540   parameters. Diurnal signal-based thermal diffusivity measurements derived from a pair of

541   1DTempProfilers inserted in sandy channel sediments for a month in 2014 have the same

542   geometric mean value of 0.11 m^2d^{-1} (Briggs et al., 2018c), and this value is used here to model

22

543 vertical groundwater discharge for all locations and data collection periods (Briggs et al., 2018c)

544 . Sub-daily groundwater discharge fluxes evaluated over similar spring/early summer time

545 periods in 2014 and 2016 show relatively stable patterns at open valley seepage zones, generally

546 <1 md^{-1} (Figure 6). At Spawn 1 and 3 seepage is stronger (2 to 3.5 md^{-1}) and more variable than

547 at open valley zones. The Darcy-based horizontal seepage estimate through the Spawn 3 bank,

548 made using the bank piezometer, is 2.3 md^{-1}, which is similar to the temperature-based seepage

549 rates at the Spawn 3 interface (Figure 6), and indicates lateral discharge through the cut bank

550 wall from a more localized groundwater flowpath. The Spawn 2 zone shows a reduced and more

551 stable discharge rate during summer 2016, and is likely impacted by a large bank slump into this

552 zone that occurred during the winter of 2016, partially filling the alcove. Seepage patterns

553 collected at Spawn 1 and 2 in late-summer 2015 show greater temporal stability, even though the

554 stream stage at the downstream USGS gage showed substantial variation. Discharge rates along

555 the inner bank wall of the scalloped bank spawn zones were consistently higher than at bed areas

556 located just a few meters away toward the channel.

557 *Streambed groundwater discharge and spawning zone pore water characterization*

558   Based on previous characterization, the Cape Cod sand and gravel aquifer generally has

559 high DO concentrations (9 - 11 mg/L), relatively dilute specific conductance (SpC, 62 µS/cm),

560 and dilute chloride concentrations (Cl$^-$, 9.3 mg/L) at depths ranging between 12 and 20 m

561 (Savoie et al., 2012). The groundwater that discharges to the Quashnet River, however, is often

562 strongly variable in all three of these parameters (Harvey et al., 2018). In June 2014, drivepoint

563 data were primarily collected in open valley seepage zones identified with FO-DTS (Figure 1);

564 these locations are suboxic to anoxic at 0.3 and 0.6 m streambed depths (Table 1). Highest

565 streambed seepage DO is found at GW330 in the tighter upstream valley section (4.6 mg/L at

566   both depths) and Spawn 3, where DO is 9.0 and 7.6 mg/L at 0.3 m and 0.6 m depths,

567   respectively (Table 1). SpC is also variable, but lowest and similar to the regional signal at

568   GW330 and Spawn 3. Note, SpC and Cl⁻ are used here to indicate aquifer flowpath

569   hydrogeochemical properties, and not suitable spawn habitat based on chemical concentration, as

570   their range is well within general brook trout tolerances.

571   Drivepoint data collected at the 0.3 m depth in June 2016, primarily around spawn zones,

572   generally show high DO and relatively low SpC at the interior of Spawn Zones 1 and 3 near the

573   cut bank (Table 1). Data collected a few meters toward the main channel from these near-bank

574   spawn locations are reduced in DO with increased SpC. The Spawn 2 data were collected at the

575   toe of the recent large sediment slump that had partially filled the alcove, and DO data are

576   suboxic at 0.3 m (3.9 mg/L) but more oxygen enriched at 0.9 m depth (7.2 mg/L) indicating the

577   potential for shallow streambed respiration that removes oxygen from discharging groundwater

578   (assuming vertical flow) in the slumped material. In contrast to the spawn zones, major open

579   valley seepage location GW1045 is nearly anoxic at all depths with SpC similar to the 2014

580   stream water profile grab samples (n=8, 101.4 +/- 1.7 µS/cm). Little difference was observed

581   between near-bank and channel positions at GW1045 (both are suboxic) even though a large

582   scalloped seepage bank feature was observed (Figure 3b).

583   The drainage-ditch grab samples generally show Cl⁻ concentrations that are lower than

584   the average 2014 channel grab samples (n=10, 19 +/- 0.4 mg/L), though the 2 most upstream

585   ditches are similar to stream water, and 2 open valley ditches are appreciably higher in Cl⁻

586   (Figure 7a). Spawn Zones 1, 2, and 3 approximate the lowest Cl⁻ concentrations observed in

587   drainage ditches, and Spawn 3 has a similar concentration to the adjacent 2016 streambank

588   piezometer in both the 2014 and 2016 data. An analogous pattern is shown in the more

24

589   widespread SpC data, with many drainage ditches and all spawn zones having concentrations

590   around 60 µS/cm, but several ditches cluster around the stream water average or higher,

591   particularly in the open valley area.

592   The shallow, discrete interval shallow pore-water samples collected with the

593   MINIPOINT system show that streambed SpC is appreciably lower than stream water, even at

594   the 0.02 m depth, at all near-bank spawn zones (Figure 8a). Conversely, the shallow channel

595   sediments at Spawn 1 and open valley seepage GW1045 approximate the stream water value for

596   SpC. DO is high and stable along the shallow profiles (to 0.14 m) at the interior of Spawn Zones

597   1 and 3, but suboxic at the Spawn 1 channel sample and Spawn 2 zones, and essentially anoxic

598   along the bank at GW1045 . Center channel pore water samples at GW1045 show moderate

599   oxygen enrichment at 0.02 m (4.6 mg/L), which may result from hyporheic mixing, as deeper

600   intervals along the same profile are nearly anoxic.

601   Underwater video collected here in the fall of 2015 indicates Quashnet River brook trout

602   clustered tightly around an approximate 1-m^2 bed area in Spawn 1 (Figure 3d, Video S1),

603   directly at the base of the sandy cut bank. During the June 2016 collection of pore-water data,

604   drivepoints were installed precisely in this area. Chemical analysis of 0.3 m depth pore water

605   shows a strong gradient from the near-bank Spawn 1 zone to the outer alcove area, with specific

606   conductance rising dramatically (70.6 to 143.9 µS/cm) and DO falling (7.28 to 4.41 mg/L)

607   (Table 1). Spawn 3 shows a similar pattern from near-bank toward main channel (60.4 to 82.1

608   µS/cm SpC; 9.11 to 1.76 mg/L DO), and Spawn 2, although complicated by the large slump

609   during the previous winter, shows an increase in SpC from 70.6 to 139.3 µS/cm from the inner to

610   outer alcove. Conversely, pore water collected at 0.3, 0.6, and 0.9 m depths in the open valley

611   seepage alcove at GW1045 (pictured in Figure 3b) are functionally anoxic with elevated SpC

612   compared to inner spawn zones, and little gradient from bank to channel.

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

614   emergent hillslope springs, characterized by relatively high DO and low SpC compared to major

615   open valley streambed seepage zones that are anoxic with higher SpC (Table 2). Additionally,

616   the stable isotopic signatures of the hillslope and Spawn zones are similar, but contrasted by the

617   lower deuterium excess metric determined for the open valley seepages. This indicates that

618   groundwater discharging through the streambed away from the hillslope shows the evaporative

619   signature of groundwater flow through lakes, and can therefore be considered regional discharge,

620   compared to locally recharged hillslope groundwaters apparently favored be trout for spawning..

621   *Visualizing streambed sediment geologic structure*
622   Radar data were collected over most of the study reach length depicted in Figure 2a, and

623   although spatial reference data were not collected for each sample point due to integrated ~~Global~~

624   global ~~Positioning~~ positioning ~~System~~ system failure, Spawn and groundwater discharge zones

625   of interest were precisely marked in the record (Figure 5). The GPR data collected along the

626   thalweg adjacent to Spawn 1 and 2 indicate a contiguous thin layer of material underlies the

627   sandy streambed that may be peat deposited over deeper sands and gravels (Figure 5a). The GPR

628   profile through open valley groundwater discharge locations GW1045 and GW1070 show the

629   strongest radar signal reflectors of anywhere along the open valley section (Figure 5b). These

630   discontinuous geologic structures are interpreted as layered sand, gravel, interspersed with

631   thicker peat deposits. Otherwise, discontinuous reflections indicative of sediment type-interfaces

632   of variable depth are observed near downstream open valley seepage zones where strongly

633   attenuated GPR signals indicate thick lenses of buried peat with high water content (Figure 5b,c).

Discussion

634 **Discussion**

635   Heat tracing reconnaissance technologies, such as FO-DTS and thermal infrared, offer an

636 efficient means to comprehensively characterize a subset of preferential groundwater discharge

637 points at the reach to watershed scale (e.g. Figure 1, Figure 4). Using the groundwater-fed

638 Quashnet River as an example, Rosenberry et al. (2016) showed that cold streambed interface

639 anomalies in summer indeed corresponded to discrete zones of particularly high groundwater

640 discharge through streambed sediments. This spatial characterization of discharge points alone is

641 not sufficient to characterize the physical and chemical drivers of niche critical cold-water

642 habitat, but can efficiently guide additional data collection, as was done here. Compared to more

643 randomly distributed streambed field parameter surveys, or larger spatial scale evaluations of net

644 groundwater discharge made with differential gaging, comprehensive spatial mapping of

645 groundwater discharges is a great advance in the context of understanding groundwater

646 dependent ecosystems. However, in fast flowing streams, FO-DTS cable placement on the

647 streambed will likely impact which specific groundwater discharge zones are captured with FO-

648 DTS, as shown here by applying cables along opposite banks through the Spawn 3 area (Figure

649 4). The largest seepage zones may have a spatial footprint that encompasses the streambed area

650 from bank to bank (e.g., the Spawn 3 cold anomaly), but a subset of more discrete seepage zones

651 are bound to be missed with a single linear cable deployment. We did not capture Spawn Zones 1

652 and 2 in early FO-DTS field efforts (Figure 1), but fish tracking indicated their importance to

653 trout spawning behavior. Therefore, in studies of niche stream habitat as influenced by

654 preferential groundwater discharge, a combination of heat tracing and biological observation

655 may be needed to both identify major discharge points and discern which points are directly used

656 by the biota of interest (e.g. brook trout).

27

657   In a study of the regional Cape Cod aquifer condition, Frimpter and Gay (1979) state that

658   groundwater is typically near DO saturation, except downgradient of peat or river bottom

659   sediments, where consumption of DO allows the mobilization of natural iron and manganese.

660   Visible observations along the open valley section, in addition to streambed sediment coring

661   (Briggs et al. 2014), revealed widespread coating of shallow streambed sediment grains with

662   metal oxides, consistent with the conceptual model of organic material influence on near-surface

663   groundwater (Figure 9). Aquifer recharge passing through upgradient groundwater flow-through

664   kettle lakes (e.g. Stoliker *et al.* 2016) may also serve to decrease the DO content of regional

665   flowpaths that discharge vertically through the bed of the Quashnet River, although we

666   hypothesize that localized peat deposits may be the primary control on both seepage zone

667   distribution and chemistry.

668   Out of the dozens of preferential groundwater discharge zones located along the lower

669   Quashnet with heat tracing, most were suboxic to anoxic (Table 1). Brook trout consistently

670   prefer three areas for fall spawning, all along meander bend cut banks into the sand and gravel

671   valley wall. Zones of locally enhanced seepage, likely controlled by subtle differences in

672   sediment hydraulic conductivity, can lead to groundwater sapping of fines, reduction in bank

673   stability, and consequent slumping of bank material into the river; this process was observed in

674   real-time at the Spawn 3 meander in February 2016 (Figure 3c). Slumping effectively forms

675   *seepage-driven* alcoves outside of the main flow and more suitable for redd placement, along

676   with a more favorable ~~course~~ coarse sand and gravel substrate (Bowerman et al., 2014; Hausle

677   and Coble, 1976; Raleigh, 1982).

678   In other systems, trout have been observed to occupy microhabitat around and within

679   groundwater discharge zones, even segregating by fish size and desirable temperature range

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680   (e.g., Figure 2.4.1.2 in Torgersen et al. 2012). Here, real-time observation and visual imagery

681   show trout clustering tightly against the bank in Spawn 3 (Figure 3d, Video S1) where pore

682   water was found to be more oxygen rich and lower in SpC. Month-long time series of vertical

683   groundwater discharge rates are reduced considerably from near-bank to near-channel at all

684   spawning zones (Figure 6), indicating in part a reduction in streambed hydraulic conductivity as

685   influenced by peat deposits under the main channel as observed in GPR data (Figure 5). The

686   combined evidence of higher near-bank vertical groundwater flux rates and DO, combined with

687   lower SpC, indicates limited interaction between shallow groundwater flowpaths and peat

688   against the meander bend cut banks. It appears that even short travel distances through organic

689   deposits toward the center channel at Spawn 1 and 2 may be sufficient to increase total dissolved

690   solids and deplete DO, as observed in other systems (e.g. Levy et al., 2016), and render

691   upwelling zones undesirable for redd construction. Therefore, near-surface channel sediments

692   may need to be specifically characterized in preferential groundwater discharge zones, as net

693   chemical reactivity over the last ~1 m of transport may dominate that along km of upgraient

694   groundwater flow through mineral soils.

695   Only where seepage was observed to emanate directly from the valley wall sands and

696   gravels, such as the newly exposed slump in Figure 3C, may groundwater discharge reliably

697   support overwinter trout egg development. These features are apparently similar to the numerous

698   cold-water alcove patches observed in another stream system by Ebersole et al. (2003). In that

699   study of preferential salmonid habitat, alcoves were often located where streams converged on

700   valley walls and were the most abundant type of discrete cold-water habitat type identified.

701   Conversely, valley wall alcoves were the least-common type of seep morphology observed along

702   the Quashnet River. It is likely that the artificial reduction in channel sinuosity along the

29

703 Quashnet River by farming practices has reduced ~~possible~~ the number of natural higher-quality

704 spawning locations.

705   Other bank alcove features with strong groundwater discharge found along the open

706 valley section (Figure 3b) were highly influenced by organic material deposition and did not

707 apparently support spawning habitat. Our research indicates that in lowland systems with

708 organic-rich floodplain sediments valley wall alcoves alone create favored brook trout spawning

709 habitat via local mineral soil-dominated groundwater discharge flowpaths as shown in

710 conceptual Figure 9. This finding might help inform future ecologically -based stream restoration

711 practices in using the natural landscape to predict desirable preferential groundwater discharge

712 points, as was recently done by Hare et al., (2017) to inform the engineering of a large-scale

713 cranberry bog restoration.

714   The pore-water SpC, Cl-, and DO data alone do not definitively show that seepage at the

715 cut bank spawn sites is derived from more localized groundwater recharge, as opposed to

716 regional groundwater that is unadulterated by buried peat lenses. However, the hydrodynamic

717 data derived from long-term vertical temperature profiling in seepage zones does offer additional

718 insight. In general, groundwater discharge rates are more variable at cut bank spawn zones than

719 in the open valley streambed zones (Figure 6), and this variability may be tied to shorter-term

720 changes in local river stage and/or water table depth, impacting the local hydraulic gradient. The

721 relatively stable patterns of open valley groundwater discharge may be controlled by the regional

722 gradient where the flowpath-length term dominates the Darcy relation, and is therefore relatively

723 insensitive to local changes in river stage and water table fluctuations. Further, the stable water

724 isotope data display evaporative signatures at the open valley streambed discharge sites,

725 indicating regional groundwater that has passed through one or more upgradient flow-through

726　lakes (Table 2). In contrast, the Spawn sites all show isotope signals that fall along the local

727　meteoric water line, and therefore likely represent recharge to the hillslopes more local to the

728　river. These localized groundwater flow systems would be expected to be less-influenced by

729　regional groundwater contamination, which is widespread in the regional Cape Cod aquifer

730　(Walter and Masterson, 2002).

731　　　Groundwater drainage-ditch data collected along the river corridor indicate low SpC/Cl⁻

732　conditions exist for the majority of ditches throughout the lower Quashnet River riparian areas

733　(Figure 7). The hillslope piezometer in sand and gravel at the down valley wall has a similar

734　chemical signature along with high DO. This similarity is further indication that low-SpC

735　groundwater discharges even to the lower portion of the river corridor, but is predominantly

736　modified chemically by travel through near-stream organics. The relic drainage ditches allow

737　discharging groundwater to effectively short circuit the valley floor peat deposits and remain

738　high in DO, similar to the natural valley wall springs and cut bank alcoves. Future restoration

739　strategies that seek to actively enhance groundwater discharge (e.g. Kurylyk et al., 2015a) may

740　consider capitalizing on this short circuit behavior, possibly by auguring through buried

741　streambed peat or movement of the stream channel toward the valley wall to create more

742　desirable brook trout aquatic habitat.

743　**Conclusions**

744　　　The three repeatedly utilized discrete spawning zone locations that have been identified

745　~~over 10 yr~~ for over a decade of observation have coupled strongly discharging groundwater with

746　high DO concentration. A conceptual diagram of the hydrogeochemical setting of spawn zones

747　vs other non-favored streambed groundwater discharge locations is shown in Figure 9. Spawn

31

748 zones are located exclusively in side alcoves of the channel created by bank slumps along

749 meanders where the river cuts into steep hillslopes along the glacial sands and gravel valley wall.

750 In the alcoves at the base of the cut banks, hillslope groundwater with high DO concentration is

751 discharged through the streambed without appreciable loss of oxygen. Just a few meters away

752 toward the main channel, however, groundwater consistently discharges at lower rates, is

753 reduced in DO, and increased in SpC. The lowest oxygen concentrations in groundwater are

754 associated with water emerging from the streambed adjacent to wide riparian areas that flank the

755 Quashnet in the open valley section of the study reach, even though groundwater discharge rates

756 were also relatively high. In the open valley, where the stream is not near the valley walls,

757 proximity to the stream bank does not seem to control seepage chemistry, and GPR data

758 indicated thick zones of discontinuous streambed peat. In this and other groundwater-dominated

759 streams that are expected to serve as climate refugia for future native trout populations,

760 hyporheic exchange will be limited by strong upward hydraulic gradient. Therefore, preferential

761 spawning habitat in such lowland valley systems may be primarily supported by discrete zones

762 of oxic groundwater upwelling at the meter to sub-meter scale as has been indicated by previous

763 work (e.g. Curry et al., 1995).

764   In systems where all groundwater discharge is universally anoxic, preferential salmonid

765 spawning zonation may be controlled by points of downwelling hyporheic water where shallow

766 sediments remain high in DO (Buffington and Tonina, 2009; Cardenas et al., 2016). However,

767 these hyporheic areas will deliver cold surface water to shallow sediments during winter, which

768 may impair overwintering brook trout eggs (French et al., 2016). Here, and in many other coastal

769 systems, groundwater temperature is expected to range approximately 10-12 $^{\circ}$C, which is an

770 ideal range for brook trout egg development (Raleigh, 1982). Points of oxic groundwater

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771 upwelling devoid of near-stream buried organics, combined with a recirculating side alcove and

772 favorable sand and gravel sediments, may provide ideal and unique groundwater seepage-

773 enabled preferential spawning habitat for native trout.

774 Stream surface or streambed interface heat tracing of groundwater discharge offers an

775 efficient means to locate discrete seepage zones, but offers only limited insight into source

776 groundwater flowpath hydraulics and geochemistry. A combined toolkit that also includes

777 spatially-informed (using heat tracing) geochemical and isotope sampling and geophysical

778 imaging can be used to trace groundwater flowpaths back into the source aquifer, and develop a

779 robust hydrogeochemical characterization. Additionally, as digital elevation models become

780 more refined and combined with infrared data derived from uUnmanned Aircraft aerial

781 Systemssystems, remote identification of relatively small features such as the seepage alcoves

782 described here should be possible. Comprehensive and process-based characterization niche

783 stream habitat can be used to guide stream ecological restoration design that directly incorporates

784 the local preferential groundwater discharge template.

785 **Acknowledgements**

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Tables

Table 1. This table lists 2014 and 2016 drivepoint pore-water chemistry data collected in major streambed groundwater discharge zones located with fiber-optic heat tracing, and in zones of observed repeat trout spawning directly along the bank and farther toward the stream center channel.

| *open valley groundwater discharges*	*0.3 m depth*		*0.6 m depth*	
	DO	SpC	DO	SpC
	mg/L	μS/cm	mg/L	μS/cm
GW330	4.6	53.8	4.6	61.3
GW880	1.4	97.7	3.4	65.1
GW1045	0.1	78.8	0.0	82.5
GW1045 (bank)	0.16	105.5	0.39	104.0
GW1045 (channel)	0.31	99.1	0.18	96.4
GW1070	0.2	100.0	0.2	89.8
GW1410	0.0	77.7	0.0	79.0
GW1470	0.1	69.1	0.0	64.3
GW2060	1.4	75.0	0.5	79.4
mean	**0.9**	**84.1**	**1.0**	**80.2**
spawning locations (channel)	*0.3 m depth*		*0.9 m depth*	
Spawn 1 channel	4.41	143.9	5.68	143.2
Spawn 2 channel	5.25	139.3	n/a	n/a
Spawn 3 channel	1.76	82.1	2.68	79.9
mean	**3.8**	**121.8**	**4.2**	**111.6**
spawning locations (bank)	*0.3 m depth*		*0.9 m depth*	
Spawn 1 bank	7.28	70.6	9.76	55.9
Spawn 2 bank	3.89	70.8	7.17	57.6
Spawn 3 bank (2016)	9.11	60.4	4.91	71.9
Spawn 3 bank (2014)	9.0	56.4	7.6 *(0.6 m)*	60.9 *(0.6 m)*
mean	**7.3**	**64.6**	**7.4**	**61.6**

40

1003   Table 2. This table lists 2017 drivepoint pore-water chemistry and stable water isotope data

1004   collected in a subset of major streambed groundwater seepage zones, zones of observed repeat

1005   trout spawning, and from springs located above the water line along the same hillslope as the

1006   meander cutbanks of Spawn 1 and Spawn 2.

location	sample depth (m)	SpC (µS/cm)	DO (mg/L)	δ^2H (‰)	$\delta^{18}O$ (‰)	d-xs $\delta^2H - 8*\delta^{18}O$
Hillslope 1	40	74.82	5.004	-51.38	-8.2	**14.22**
Hillslope 2	44	60.59	9.318	-51.81	-8.73	**18.03**
Spawn 1	20	72.45	6.853	-48.9	-7.9	**14.3**
Spawn 2	20	51.75	5.419	-48.2	-7.95	**15.4**
Spawn 3	20	42.62	9.054	-44.32	-7.33	**14.32**
GW1045	20	109.8	0.043	-34.03	-4.93	**5.41**
GW1140	20	103.4	0.043	-32.56	-4.8	**5.84**
GW1470	20	97.68	0.04	-33.05	-4.72	**4.71**

1007

1008

1009 **Figure List**
1010
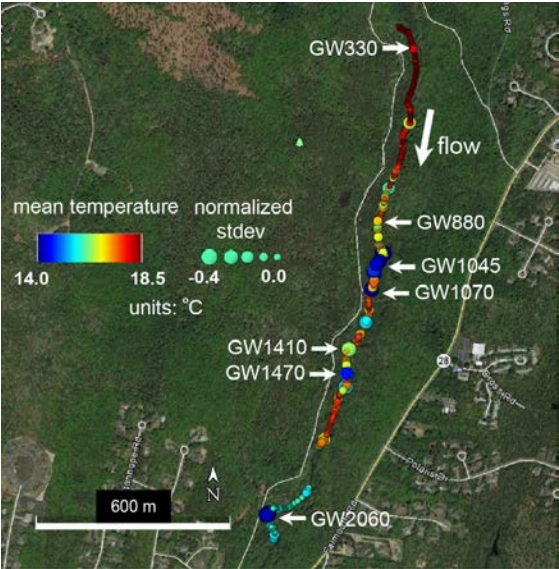
1011
1012 Figure 1. Fiber-optic distributed temperature data collected along the stream channel sediment-
1013 water interface over two days in July 2013 are summarized here using mean temperature (color)
1014 and temperature standard deviation normalized to known non-seepage locations (size). Locations
1015 of reduced mean temperature and temperature standard deviation can indicate zones of
1016 preferential groundwater upwelling. A subset of these apparent upwelling zones (labeled "GW"
1017 followed by the distance from upper reach boundary in meters) with varied thermal statistics
1018 were chosen for direct pore water sampling and quantitative seepage measurements. This Figure
1019 was modified from Rosenberry et al. (2016).
1020
1021
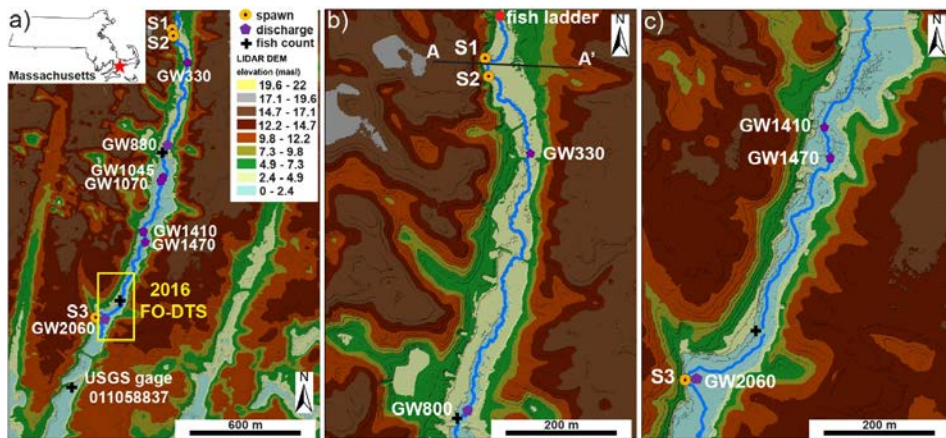
Figure 2. Lidar elevation data show the linear valley terrain of: a) The lower the Quashnet River study reach, as shown in panel (a) with Spawn (S1, S2, S3) locations and major open valley seepage zones identified. All seepage zones are labeled as downstream distance from the stream crossing/fish ladder located at the upper extent of this image. The zoomed in view of pPanel (b) shows the tighter more narrow upper valley zone where Spawn 1 and 2 are located at the base of a steep cut bank, and the topographic transect of Figure 9 (A-A')1 is noted. Finally, pPanel (c) displays the lower open valley reach where Spawn 3 is located along a major cut bank.

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1031
1032 Figure 3. Several representative images of specific spawn zones and groundwater discharge
1033 zones were Images collected in February 2016 a) t. The cut bank alcove at Spawn 1 is shown in
1034 panel (a), while the b) open valley seepage zone GW1045 is shown in (b), andand the c) fresh
1035 cut bank slumping and visible seepage at Spawn 3 is shown in (c). Panel d) is an image from the
1036 underwater video collected in fall 2015 of spawning trout in the Spawn 1 alcove pictured in
1037 panel a),Underwater imagery collected at the Spawn 1 zone in fall 2015 is displayed in panel (d),
1038 showing showing several fish clustered around the sandy zone directly at the base of the cut bank
1039 where pore water samples were obtained.
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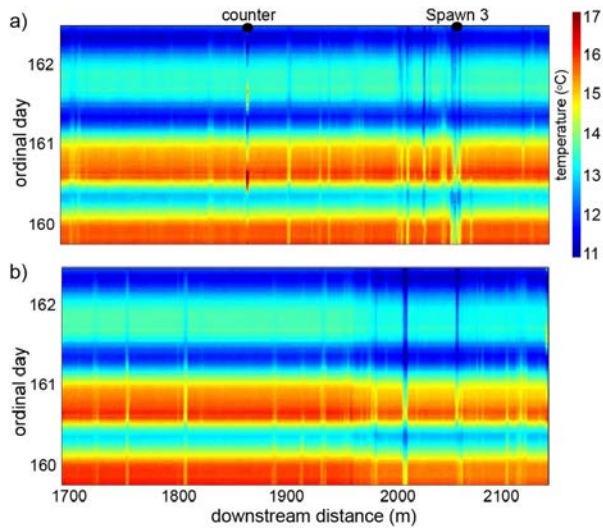
1043
1044 Figure 4. Fiber-optic-distributed temperature data collected from approximate channel distance
1045 1700 to 2160 m along (a) the downstream right bank through the Spawn 3 meander bend area
1046 (see Figure 2c 2a for location), and (b) the downstream left bank. The persistent vertical bands of
1047 relatively cool temperatures indicate discrete groundwater discharge. Some larger zones display
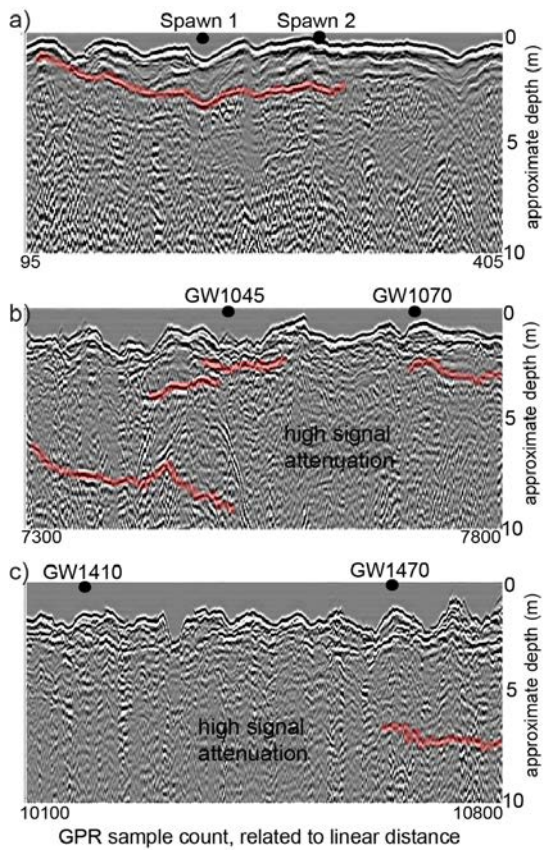1048 a thermal signature on both bank cables, while smaller discharges may be bank-specific.
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1051
1052 Figure 5. ~~Quashnet River thalweg g~~These images show ground penetrating radar profiles ~~were~~
1053 collected down the center of the river channel to indicate peat and sand/gravel layering in the
1054 streambed. Stronger apparent radar reflectors are highlighted in red, and likely indicate sediment
1055 layer boundaries (e.g. sand/gravel vs. peat). Spawn and groundwater discharge locations were
1056 directly marked in the radar data stream during collection and are shown for each sub-reach
1057 panel. ~~in the vicinity of: a) Spawn 1 and 2; b) open valley seepage GW1045 and GW1070; and~~
1058 ~~c) open valley seepage GW1410 and GW1470. Stronger apparent reflectors are highlighted in~~
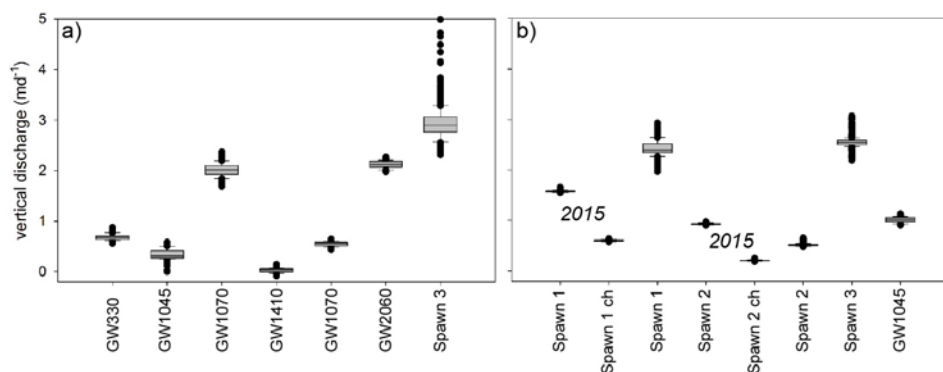1059 ~~red, and likely indicate sediment layer boundaries (e.g. sand/gravel and peat).~~
1060
1061

Figure 6. ~~Panel a) displays s~~Summary box plots of sub-daily ~~(June 11 to July 13, 2014)~~ vertical groundwater discharge rate_s modeled_ ~~estimates~~ for the open valley _groundwater_ discharge and Spawn 3 bank locations _for the 11 June to 13 July 2014 period are shown in panel (a)._ _Additionally, p_~~P~~anel (b) shows ~~vertical~~ _displays_ discharge rates collected _in Spawn and GW1045 locations directly_ against the _cut_ banks_,_ and _further out_ toward_s_ the channel _(indicated by "ch")_ _for the_ ~~at Spawn zones 1 and 2~~ _21_ ~~(~~August 21 to _13_ September ~~13,~~ 2015~~);~~ _and_ ~~and from locations against the bank in all three Spawn locations and GW1045 (June~~ _5_ June to ~~July~~ _9 July,_ 2016 _periods._ ~~).~~
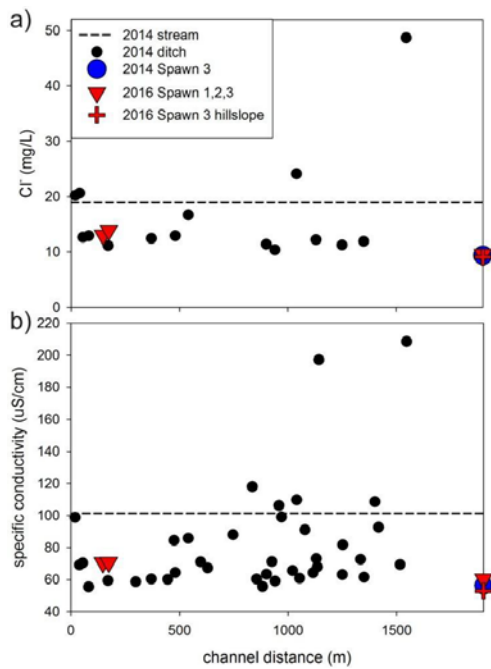
Figure 7. Drainage ditch chemistry throughout the lower Quashnet showing (a) Cl⁻, and b) specific conductance, collected in June 2014 just above the confluence with the main channel. Data are plotted as distance from the upper flood control structure in the narrow valley reach and compared to groundwater seepage data collected in preferential spawning locations and a hillslope piezometer.
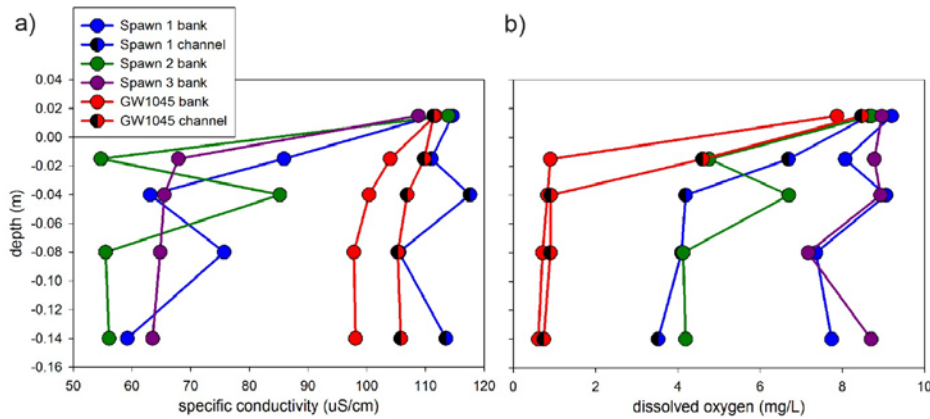
48

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.
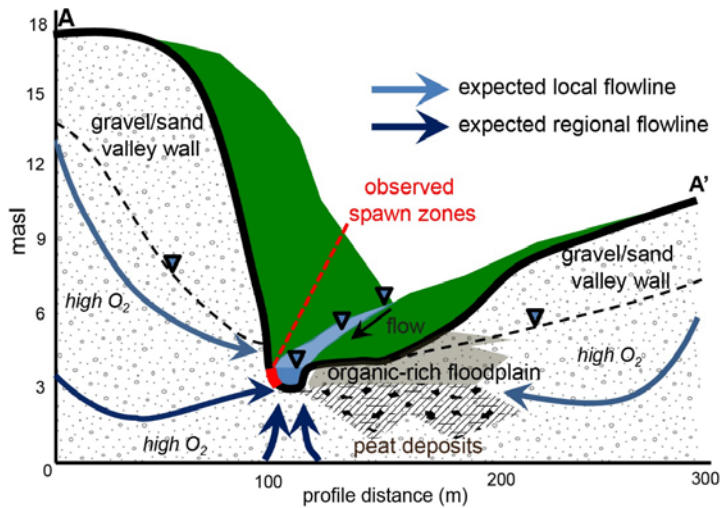

Figure 9. ~~Based on previously published USGS work (e.g. Modica, 1999; Winter et al., 1998), the conceptual model in panel a) displays how groundwater discharge to lowland streams is expected to include locally sourced lateral groundwater discharge through valley wall features and more regionally sourced groundwater discharge vertically through the streambed.~~ This conceptual model shows how valley wall cutbank discharge zones are likely sourced by locally recharged hillslope groundwaters that avoid substantial interaction with valley floor organic material. The discharging groundwater remains oxygen-rich, therefore supporting trout spawning activity along discrete streambed sections at the meter scale. The topographic profile shown here

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1105   (A-A') is derived directly from lidar data ~~in the vicinity of observed preferential brook trout~~
1106   ~~spawning habitat shown in Figure 2.~~collected normal to the stream through the Spawn 1 zone, as
1107   geolocated in Figure 2b. ~~In contrast to the sand and gravel valley walls, multiple methodologies~~
1108   ~~used for this study indicate wider valley zone sediments to be rich in organic material, including~~
1109   ~~buried peat deposits, consistent with known regional geology.~~
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1111   **Supplemental**
1112
1113   Supplemental Video S1. Underwater video of brook trout spawning in the fall of 2015 (still
1114   image Figure 3d).