5 Tracking repeat-spawning anadromous fish over multiple migrations reveals individual repeatability, tagging effects and environmental factors influence barrier passage

5.1 Abstract

1) Understanding the impacts of anthropogenic barriers on anadromous fish migration is an important step in planning and predicting the consequences of river reconnection. In addition, it is necessary to understand potential biases that occur when using biotelemetry to estimate barrier impacts on fish migration.

2) This study focused on the freshwater spawning migration of twaite shad *Alosa fallax,* a repeat-spawning anadromous fish, in a heavily fragmented river catchment over three study years. Passive acoustic telemetry was applied to 184 individuals to assess the impact of multiple weirs in the River Severn catchment (western Britain), on their upstream distribution, the factors affecting their approach to weirs and the individual and environmental factors affecting weir passage rates. Most fish (94%) were tagged with 3-year transmitters, enabling an assessment of barrier passage by returning individuals tagged in previous years versus newly tagged fish, as well the impact of previous passage experience on barrier passage.

3) The proportion of fish that approached and passed barriers varied between years and weirs, with higher proportions approaching and passing the tidally affected weirs further downstream in the catchment. Of the individuals tagged with 3-year transmitters, 71 (57%) returned for a second year, and the proportion of these that passed a major navigation weir in the lower catchment (S2) was higher in 2019 and 2020 (82%, 65%) than newly tagged individuals in 2018 and 2019 (40%, 17%). Median cumulative passage times (lower quartile-upper quartile) at weirs was 4.6 (1.8 - 9.2) days and represented 18% of the 25 (16 - 32) days total time in fresh water recorded by emigrating tagged individuals.

4) Time-to-event analysis of passage of weir S2 revealed that returning individuals had significantly higher passage rates than newly tagged individuals,

and amongst returning individuals, passage success in the previous year was significantly associated with higher passage rates. In addition, increasing temperature and higher river levels had significant positive impacts on the rate of passage.

5) This study demonstrated that navigation weirs inhibited the freshwater spawning migration of a threatened anadromous species by delaying or preventing upstream passage, with passage strongly influenced by environmental factors. Higher weir passage rates by returning versus newly tagged individuals suggests that reliance on the latter in barrier impact assessments could result in conservative estimates of passage, while higher passage rates of previously successful versus unsuccessful individuals suggests a conserved motivation and/ or inherent ability to pass barriers.

5.2 Introduction

There are now few rivers that remain free-flowing along their entire length, particularly in developed regions (Jones et al., 2019; Belletti et al., 2020). Anthropogenic fragmentation of riverine ecosystems occurs primarily through river-regulation structures, such as dams and weirs, which are constructed for a variety of purposes including power generation and to enable navigation (Grill et al., 2019). A major ecological impact of river fragmentation is its disruption of migrations of diadromous fishes (Hall, Jordaan & Frisk, 2011; Birnie-Gauvin, Tummers, et al., 2017). Severe and widespread declines in diadromous fish populations have been recorded in recent decades, which have been largely attributed to habitat fragmentation along their migratory pathways that impede their access to upstream spawning grounds (Limburg & Waldman, 2009).

Man-made structures in rivers disrupt the migration of diadromous fishes by acting as physical impediments that prevent or delay access to optimal spawning habitat (Lundqvist et al., 2008; Castro-Santos, Shi & Haro, 2017; Newton et al., 2018). Delays at barriers can increase predation risk and there are negative energetic consequences associated with locating and negotiating passage routes, especially when there are multiple passage attempts (Castro-Santos &

Letcher, 2010; Nyqvist et al., 2017). Where rivers contain multiple barriers, the effects of sequential barriers can be cumulative (Keefer, Boggs, et al., 2013; Castro-Santos, Shi & Haro, 2017) and so restoring connectivity in these rivers requires prioritisation of the structures according to the extent of their relative impacts (Kocovsky, Ross & Dropkin, 2009; Nunn & Cowx, 2012; King et al., 2017). For adult anadromous fish, this knowledge can be gained through telemetry studies that track a subset of the upstream migrants to determine, for example, the proportion of fish passing each barrier, the extent of the delay at barriers and their final upstream distribution (Thorstad et al., 2008; Keefer, Boggs, et al., 2013).

Barriers to migrating anadromous fish are often semi-permeable, where they prevent a proportion of migrants from progressing, and/or subject migrants to a delay (Castro-Santos, Shi & Haro, 2017; Newton et al., 2018). As migration and thus barrier passage are time-limited processes, telemetry data analyses often adopt a rates-based approach that enable assessments of the impacts of time-varying and time-constant covariates on passage rates (Castro-Santos & Haro, 2003). Such studies have revealed that environmental factors, such as river discharge and water temperature, can have significant effects on barrier passage rates, with higher discharge and temperatures resulting in higher passage rates (Nyqvist et al., 2017; Harbicht et al., 2018). In addition, individual factors such as body size, shape and condition, can affect barrier passage rates (Keefer et al., 2009; Nau et al., 2017; Goerig et al., 2020), while behavioural and social factors can also be important, such as collective navigation and the ability to explore for alternative passage routes (Kirk & Caudill, 2017; Okasaki et al., 2020). Although iteroparous fishes that spawn multiple times in their natal river across their lifecycle potentially encounter the same barriers on multiple occasions, the effect of these previous encounters on their barrier passage is poorly understood (Nau et al., 2017). However, assessments of how individuals perform at the same barriers in different migrations should increase understandings of how interactions of individual and environmental factors influence passage, and help predict colonisation of new areas following barrier passage remediation (Pess et al., 2014). These assessments could also indicate whether potential biases are incurred in data that are reliant on only newly tagged fish through comparing passage rates between their year of tagging and their subsequent return (Nau et al., 2017).

An anadromous fish that is becoming increasingly threatened across its range is the twaite shad *Alosa fallax*, which is distributed across the north-eastern Atlantic and Mediterranean (Aprahamian et al., 2003a). Recent declines and extirpations of their populations in European rivers have been attributed to pollution, overfishing and man-made structures that act as barriers to their upstream spawning migration (de Groot, 1990; Aprahamian et al., 2003a; Antognazza et al., 2019). In the northern part of their range, they are highly iteroparous, with fish that have previously spawned often representing over 50 % of all migrants (Aprahamian et al., 2003b). Although they are sensitive to handling and sedation, recent advances in surgical tagging protocols have enabled internal transmitter implantation, facilitating assessments of individual movements across successive migrations (including evaluating individual repeatability in barrier approach and passage), provided the implanted tags have a long battery life (Bolland et al., 2019b). This has also provided insights into the marine phase of the lifecycle of shad that use the highly fragmented lower River Severn catchment, western Britain, for their spawning (Davies et al., 2020/Chapter 4).

Here, the aim was to assess the impacts of weirs as barriers to the spawning migrations of twaite shad. Through application of long-life acoustic tags, multiple freshwater spawning migrations of twaite shad were tracked in the lower River Severn catchment, enabling completion of the following objectives: 1) estimate the impacts of anthropogenic barriers on twaite shad upstream migrations, including the proportion of upstream migrants passing barriers and migration delay imposed on individuals; 2) determine the upstream extent of twaite shad spawning migration with respect to anthropogenic barriers and major tributaries, and the factors affecting approach of weirs; and 3) determine the individuals and environmental factors influencing passage rates of artificial barriers by twaite shad, including comparisons of passage rates of newly tagged versus returning individuals, and previously successful versus unsuccessful individuals.

5.3 Methods

5.3.1 Study duration and area

The study assessed the upstream spawning migrations of twaite shad in the River Severn in 2018, 2019 and 2020, which tend to commence in April and conclude in June (Antognazza et al. 2019). The Severn is the longest river in Great Britain, rising in mid-Wales and flowing for 354 km before discharging into the Bristol Channel, and has a drainage area of 11420 km^2 (Durand et al., 2014). The study area in the lower river catchment includes confluences with two major tributaries, the River Teme and River Avon, and 8 major weirs (four on the main river channel, and two on each of the lower reaches of the River Teme and River Avon) that result in high fragmentation (Figure 5.1, Table 5.1). The normal tidal limit is at Maisemore (hereafter S1a) and Llanthony Weirs (S1b) on the western and eastern branches of the river respectively (Figure 5.1), although large spring tides can penetrate the river up to Upper Lode Weir (hereafter S2). Between the spawning migrations of 2018 and 2019, the two weirs on the River Teme (T1, T2; Figure 5.1) were modified by lowering (T1) and reducing the approach gradient by installing a rock ramp (T2). With the exception of S2, which featured a notch fish pass, there were no fish-passage structures on weirs in the Severn during the study period. Environmental data for the study reach were obtained by request from the Environment Agency's gauging stations at Saxon's Lode (temperature, River Severn, approximately 3 km upstream of S2 (Figure 5.1), Ashleworth (river level, River Severn, approximately 10 km downstream of S2), and T2 (discharge and temperature, River Teme) (Figure 5.1). All environmental data were collected at 15-minute intervals.

Table 5.1: Locations of study weirs in the River Severn catchment, which were used to assess the impacts of weirs and factors affecting approach and passage on the upstream migration of acoustic-tagged twaite shad

Weir code	Name	River	Location, decimal degrees	Distance from normal tidal limit, rkm	Original function
S1a	Maisemore Weir	Severn (West Channel)	51.89318, -2.26574	0	Navigation
S1b	Llanthony Weir	Severn (East Channel)	51.86227 -2.26028	0	Navigation
S2	Upper Lode Weir	Severn	51.99346, -2.17407	16	Navigation
S3	Diglis Weir	Severn	52.17926, -2.22597	42	Navigation
T1	Powick Weir	Teme	52.16975, -2.24712	44	Flow regulation
T2	Knightwick Weir	Teme	52.19908, -2.38940	60	Flow regulation
A1	Abbey Mill Weir	Avon	51.99133, -2.16325	16	Flow regulation
A2	Stanchards Pit Weir	Avon	51.99837, -2.15561	18	Flow regulation

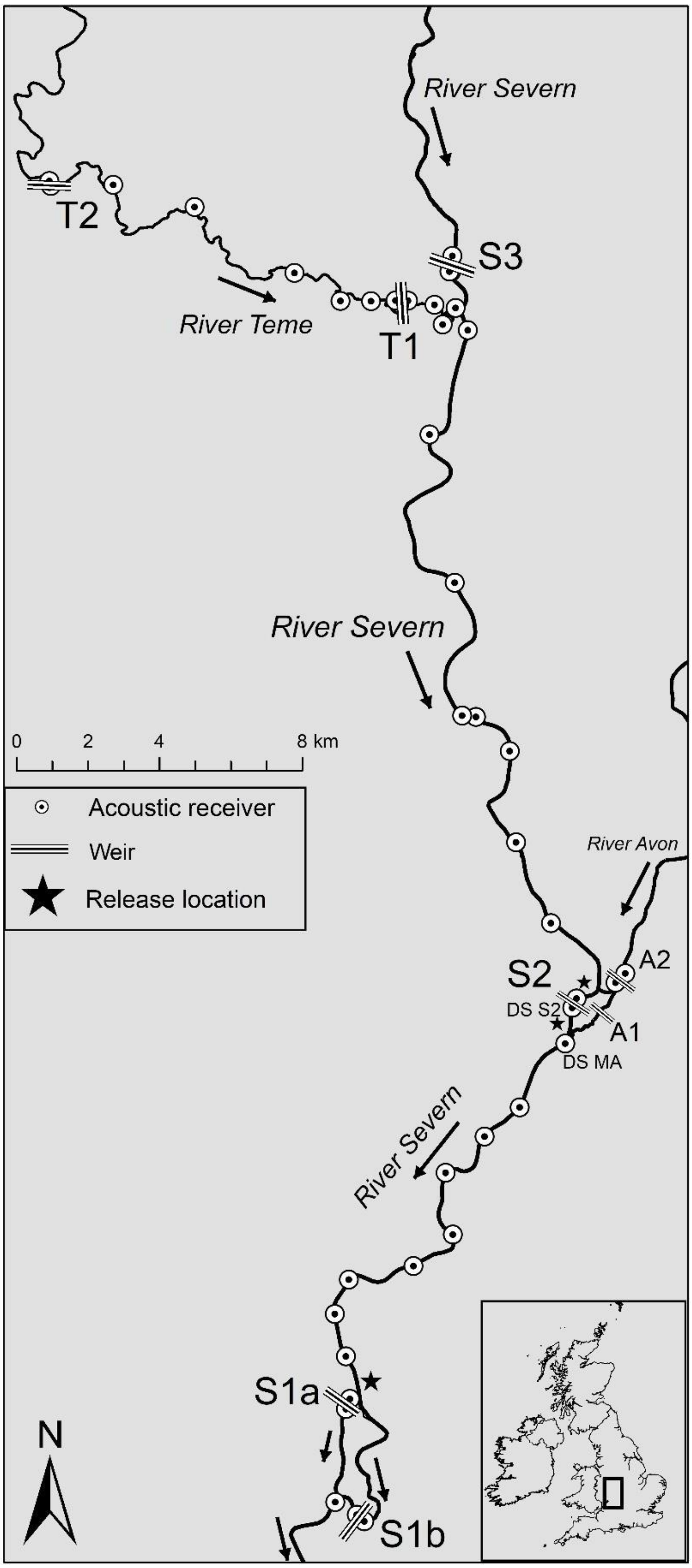

Figure 5.1: The River Severn catchment study area, including locations of release of acoustic-tagged twaite shad (black star), weirs (bars) and acoustic receivers (circles) in the rivers Severn, Teme and Avon, UK. The weir codes are as in Table 5.1. The black arrows denote the direction of the flow.

5.3.2 Fish capture, tagging and release

At the commencement of their migration season in early-mid May 2018 and 2019, upstream-migrating adult twaite shad (referred to as ‘shad’ in methods and results) were captured by rod-and-line angling in the weir pools of S1a and S2. In addition, shad were captured at S2 using a trap positioned at the upstream exit of the ‘notch’ fish pass. Following their anaesthesia (Ethyl 3-aminobenzoate methanesulfonate; MS-222), all fish were weighed (to 10g), measured (fork length, nearest mm) and approximately three scales removed for analysis of spawning history. These scales were analysed subsequently to determine their number of spawning-marks (and so their migration history) using a projecting microscope (x48 magnification) (Baglinière et al., 2001). Following the collection of their biometric data, the shad were surgically tagged with 69 kHz, Vemco V9 acoustic transmitters (vemco.com), using the tagging protocol of Bolland et al. (2019), and following ethical review and according to UK Home Office project licence PD6C17B56. A total of 184 shad were tagged over the two years (Table 5.2), of which 173 were tagged with programmed long-life acoustic transmitters. At the end of June, these transmitters were programmed to switch from a randomized 1-minute pulse interval (minimum interval between acoustic pulses 30 seconds, maximum interval 90 seconds) to a 10-minute pulse interval until April the following year, when they were programmed to switch back to their randomized 1-minute pulse interval. This was to increase the battery life of the transmitters to approximately three years, so potentially enabling the tracking of three consecutive spawning migrations of tagged individuals.

At S1a, the newly tagged shad were released approximately 100 m upstream of the weir (Figure 5.1) in order to quantify approach and passage at the next weir (S2) (Table 5.2). At S2, the majority of captured shad were released upstream of the weir in order to study the extent of their onward migration in the main river and then the impact of the subsequent weirs in the Rivers Severn, Teme and Avon. Some additional rod and trap caught fish were released downstream of S2, to increase the sample size of shad that were used to assess passage at this structure.

Table 5.2: Summary metrics for acoustic tagged twaite shad *Alosa fallax* captured over two years in the River Severn

Year	Capture location	Capture method	Release location	*n*	Length ± SE, mm	Weight ± SE, g
2018	S1a	Angling	Upstream S1a	20	365.9 ± 5.6	653.8 ± 33.2
	S2	Angling	Downstream S2	10	375.4 ± 6.5	645.0 ± 33.7
	S2	Angling	Upstream S2	24	339.8 ± 6.5	479.2 ± 29
	S2	Trap	Downstream S2	8	357.6 ± 9.9	559.4 ± 64.6
	S2	Trap	Upstream S2	22	376.4 ± 3.6	736.4 ± 24
2019	S1a	Angling	Upstream S1a	50	350.9 ± 6.1	617.5 ± 36.1
	S2	Trap	Upstream S2	50	376.9 ± 5.4	776.5 ± 35.3
Total				184	362.8 ± 2.7	659.8 ± 16.8

5.3.3 Acoustic array

Prior to the commencement of each spawning migration period, an array of Vemco acoustic receivers (VR2-W and VR2-Tx, www.innovasea.com) was installed throughout the study area (Table 5.1; Figure 5.1). The furthest downstream receiver in the array (51.8347, -2.2901; Figure 5.1) was located in the estuary, 8 km downstream of the tidal limit, at the approximate summer limit of saltwater incursion into the river (Bassindale, 1943). Receivers were deployed upstream and downstream of each navigation weir on the main channel and each flow-regulation weirs on the rivers Teme, Avon and Mill Avon, with additional receivers deployed in unobstructed reaches between weirs (Table 5.1; Figure 5.1). Although no shad were tagged in 2020 due to Covid-19 restrictions, the receiver array was installed to enable tracking of returning fish tagged in previous years. Receivers were anchored on steel fencing pins driven into the riverbed. In the River Teme, which featured sections of fast-flowing riffle, receivers were deployed in slower-flowing pools to maximise detection distance. In each tracking year, data were downloaded from receivers every two weeks until no further movements were detected. Range tests revealed that 100 % of test tag transmissions were detected a minimum of 100 m away from receivers in the River Severn, and a minimum of 50 m away from receivers in River Teme. In all cases, detection range was greater than river width at receiver deployment location. Detection efficiency calculations (using three sequential receivers to determine the efficiency of the middle receiver) revealed that missed detections accounted for less than 0.1 % of shad movements between receivers.

5.3.4 Data analysis

All statistical analyses were conducted using R statistical software (version 4.0.2, R Core Team, 2020). Initially, emigration and return rates were calculated for shad released in each tracking year, as well as for returning shad in each subsequent year. Shad were classed as having emigrated from the river if their final detection location was the most downstream receiver in the array (Figure 5.1) and they were classed as returning if they were detected moving upstream into the array in subsequent years.

To understand the relative impacts of weirs on upstream-migrating shad, the following key approach and passage summary metrics were calculated for each weir in the study area: *n* available, *n* approached, per cent approach, *n* passed, per cent passage and passage time (Table 5.3). These metrics were calculated separately for each of the study years, and for newly tagged versus returning individuals. To then understand the overall impact of weir on the upstream migration of tagged individuals, the following summary metrics were calculated for each individual in each year: upstream extent, total passage time and delay proportion (Table 5.3).

Table 5.3: Definition of metrics used to quantify approach and passage of weirs in River Severn catchment by acoustic-tagged twaite shad, and the impacts of weirs on individual migration

Metric	**Definition**	**Quantified for:**
n available	The number of fish detected moving upstream in the unobstructed reach downstream of the study weir	Each weir
n approached	The number of upstream-moving fish that were detected on the receiver immediately downstream of a weir	Each weir
Per cent approach, %	The proportion of *n* available fish that approached a weir	Each weir
n passed	The number of fish approaching a weir that were subsequently detected on an upstream receiver	Each weir
Per cent passage, %	The proportion of approaching fish that passed a weir	Each weir
Passage time, days	Time between the first detection on the downstream receiver at a weir and first detection on an upstream receiver	Each weir
Upstream extent	The furthest upstream location that a fish was detected within the catchment	Each individual
Total passage time, days	Sum total of passage times recorded at all weirs	Each individual
Delay proportion, %	Total passage time as a proportion of the time between first and last detection in the array	Each individual

5.3.5 Factors affecting approach of weirs

The individual factors affecting weir approach by newly tagged and returning shad were tested using binomial generalised linear mixed models (GLMMs) in the R package *lme4,* and generalised linear models (GLMs) in base R. Individuals that were available to approach S2 and/or S3/T1 were categorised as either approaching (1) or non-approaching (0). Two sets of models were constructed to test the effects of individual covariates on approach likelihood. The first model set tested whether tagging status (newly tagged versus returner) affected the likelihood of weir approach, using GLMMs. These models included the approach classification (0/1) for fish that provided two years of approach data at a weir. Additional individual covariates were body length and spawning history (number of previous spawning events indicated by scale analysis). A fixed effect of weir was also included to test whether approach likelihood of individuals that were available to approach S2 differed from approach likelihood of those available to approach S3/T1. To account for repeated measures from the same individuals, a random effect of individual fish id was included in the models.

The second model set tested whether approach of S3 and/or T1 in the previous year affected the subsequent likelihood of approach of either weir for returning fish, using GLMs. These models included the approach classification (0/1) of returning individuals with known approach classifications in the previous year. Additional individual covariates were body length and spawning history. Approach of S2 was not included in this model, due to high approach rates by returning individuals at this weir.

Models containing all possible combinations of covariates without interactions were tested and ranked according to AICc; models within 2 AICc of the top-ranked model were considered to have strong support (Burnham & Anderson, 2002), unless they were a more complex version of a nested model with lower AICc (Richards, Whittingham & Stephens, 2011).

5.3.6 Factors influencing passage rates of weirs

The factors influencing passage rates of newly tagged and returning shad were tested using time-to-event analysis (Castro-Santos & Haro, 2003; Goerig et al., 2020). This analysis measured the relative effects of individual and time-varying covariates on passage rates at S2 (Figure 5.1), as this weir had the largest sample size of approach and passage over the three tracking years. Shad entered the ‘risk set’ (the set of individuals to pass) when they were detected on the receiver immediately downstream of S2 during an upstream approach (Figure 5.1). Individuals remained in the risk set until their retreat downstream (confirmed by detection on receiver approximately 1 km downstream of S2 (Figure 5.1)) or their passage over the weir. Mixed effects cox models of passage rate, incorporating individual and environmental fixed effects and a random effect (shad i.d.), were constructed using the package *coxme* in R (R Core Team, 2020; Therneau, 2020). The random effect accounted for statistical dependence among repeated approach and passage from the same fish in different years (Therneau, Grambsch & Pankratz, 2003).

During data preparation, raw detection data for each shad were converted into 15-min observations of location, defined as the location of last detection, and observations of movements between receivers. Approach observations occurring at the receiver immediately downstream of S2, and passage observations (first detection upstream), were selected. These observations were then associated with individual metadata (body length, spawning history, previous success) and environmental data. Environmental covariates were downstream river level (m), water temperature (^{o}C) and diel period (as day/night, based on time of sunset and sunrise at weir S2, using the *maptools* package (Bivand & Lewin-Koh, 2019)). Individual body length (cm), spawning history (*n* previous spawning events, grouped into 0, 1, 2+) were also included as covariates. Shad that passed the weir were censored from the model dataset at the time of passage, and non-passing individuals following their final upstream approach.

Following data preparation, two model datasets were created to test specific factors relating to the tagging status and previous experience of individual tagged shad on passage rates at S2. Dataset 1 enabled testing of tagging status (newly tagged versus returning shad) on passage rates, and so contained approach and passage events for acoustic-tagged shad released downstream of S2 in 2018 and 2019 that also returned to the weir following year, i.e. 2019 and 2020. Dataset 2 enabled testing of the impact of previous success at passing weir S2 during the first year at liberty (2018 and 2019) on subsequent passage rates in the return year (2019 and 2020, respectively), so contained approach and passage events for returning acoustic-tagged shad with known passage (successful or unsuccessful) during their first year at liberty.

To analyse these two datasets, initial data exploration assessed collinearity between covariates (Zuur, Ieno & Elphick, 2010). Model selection was then conducted as per the GLMMs. The assumption of proportional hazards in the top-ranked cox models was assessed by visual inspection of Schoenfeld residuals to confirm a horizontal slope for each covariate (Schoenfeld, 1982). Covariate effects from the top-ranked model were presented as hazard ratios (HR), which represent the effect on passage rates of increasing the value of continuous covariates by one unit (e.g. by 1 m for river level) or by changing the value of a categorical covariate. Survival curves for categorical predictive variables, and representative levels of continuous predictive variables, were plotted using the R package *ggsurvplot.*

5.4 Results

5.4.1 Summary of emigration and return

Of the 173 shad tagged with long-life acoustic transmitters in 2018 and 2019, 125 (72 %) emigrated from the river (Table 5.4). Of these emigrating fish, 71 (57 %) were subsequently detected returning to the River Severn for a second year, and of these 53 (75 %) emigrated for a second time. Emigration rates were similar between newly tagged fish and returning fish in each year, and return rates were the same (57%) for newly tagged fish that emigrated in 2018 and 2019 (Table 5.4). Of the 73 fish tagged in 2018, 7(10 %) returned for a third year in 2020.

Table 5.4: Summary of emigration and return rates by twaite shad tagged with 3-year acoustic transmitters in 2018 and 2019

Year 1			Year 2		Year 3	
Year	*n* tagged	*n* emigrated (% of tagged)	*n* returned (% of emigrated)	*n* emigrated (% of returned)	*n* returned (% of emigrated)	*n* emigrated (% of returned)
2018	73	58 (79%)	33 (57%)	24 (73%)	7 (29%)	4 (57%)
2019	100	67 (67%)	38 (57%)	29 (76%)	NA	NA
Total	173	125 (72%)	71 (57%)	53 (75%)	NA	NA

5.4.2 Weir approach, passage and passage time

The percentage of shad that approached and passed weirs in the River Severn catchment varied spatially (between weirs), temporally (between years), and also between newly tagged and returning fish (Table 5.5). At S1a/b, the first weirs encountered by upstream-migrating shad, the combined per cent approach and passage of returning individuals at these structures were very high (98-100 %) in 2019 and 2020 (Table 5.5). Of those that moved upstream of S1a/b, per cent approach of the next weir S2 was high in each tracking year, particularly for returning individuals (98-100%) relative to newly tagged individuals (91-93%) (Table 5.5). Per cent passage of S2 varied between tracking years and tagging status, being lowest for newly tagged individuals in 2019 (17 %) and highest for returning individuals in 2019 (82 %) (Table 5.5). When compared with passage of weirs further downstream, passage rates of S3 were always low (Table 5.5). At T1, passage was 0 % in 2018 (*n* = 18), but following its modification in late 2018, passage rates increased to 50 % in 2019 (*n* =18), which included passage by both newly tagged and returning individuals, and 67 % in 2020 (*n* = 3) (Table 5.5). Of those shad that moved upstream of T1, few approached the next weir, T2, and no shad passed A2 in any year (Table 5.5).

Table 5.5: Summary of weir passage metrics for acoustic tagged twaite shad migrating upstream in the River Severn catchment in 2018, 2019 and 2020.

Weir	Year	Fish status	n available	n approached (% of available)	n passed (% of approached)	Median passage time, days (LQ-UQ)
S1a/S1b	2018	Newly tagged	NA	NA	NA	NA
	2019	Newly tagged	NA	NA	NA	NA
	2019	Returning	33	33 (100%)	33 (100%)	1.0 (0.4-3.9)
	2020	Returning	45	44 (98%)	44 (100%)	1.5 (1.0-2.8)
S2	2018	Newly tagged	33	30 (91%)	12 (40%)	5.9 (5.0-6.2)
	2019	Newly tagged	45	42 (93%)	7 (17%)	6.2 (2.8-33.0)
	2019	Returning	33	33 (100%)	27 (82%)	1.8 (1.1-3.4)
	2020	Returning	44	43 (98%)	28 (65%)	1.9 (1.3-4.7)
S3	2018	Newly tagged	57	29 (51%)	0 (0%)	NA
	2019	Newly tagged	56	30 (54%)	1 (3%)	21.0 (NA)
	2019	Returning	27	13 (48%)	2 (15%)	25.8 (24.6-27.1)
	2020	Returning	28	19 (68%)	0 (0%)	NA
T1	2018	Newly tagged	57	18 (32%)	0 (0%)	NA
	2019	Newly tagged	27	11 (41%)	6 (55%)	1.1 (1.1-3.8)
	2019	Returning	56	7 (13%)	3 (43%)	0.0 (0.0-0.5)
	2020	Returning	28	3 (11%)	2 (67%)	0.4 (0.3-0.5)

Weir	Year	Fish status	n available	n approached (% of available)	n passed (% of approached)	Median passage time, days (LQ-UQ)
T2	2018	Newly tagged	0	0 (NA)	0 (NA)	NA
	2019	Newly tagged	6	1 (17%)	1 (100%)	NA[1]
	2019	Returning	3	1 (33%)	1 (100%)	NA[1]
	2020	Returning	2	1 (50%)	0 (0%)	NA
A2	2018	Newly tagged	57	21 (37%)	0 (0%)	NA
	2019	Newly tagged	27	6 (22%)	0 (0%)	NA
	2019	Returning	56	10 (18%)	0 (0%)	NA
	2020	Returning	28	12 (43%)	0 (0%)	NA

[1]Passage times unavailable due to missed detections on downstream receiver

Passage times at S2 were the longest of the weirs where at least 10 passages occurred (Table 5.5); passage time also varied between years and tagging status, being longest for newly tagged fish in 2019 (median passage time (LQ-UQ) = 6.2 (2.8-33) days), and shortest for returning individuals in 2019 (1.8 (1.1-3.4) days) (Table 5.5). Median total passage times at weirs of 4.6 days (1.8 - 9.2) represented a delay proportion of 18 % of total time-at-large (25 (16-32) days) for individuals that were tracked re-entering the estuary after their freshwater migration.

Of the movements recorded upstream of S1a/b (*n* individuals = 114; *n* upstream movements = 152), 94 % resulted in an approach of S2, with the others reaching their upstream extent between 1 and 4 rkm downstream of S2 (Figure 5.2). Of the upstream movements recorded upstream of S2 (*n* individuals = 127; *n* upstream movements = 164), 63 % approached S3 and/or T1, and upstream extents for non-approaching fish were concentrated around the lower River Teme and its confluence with the Severn (19 %, Figure 5.2), with a further 19 % reaching an upstream extent within the 24 rkm section of the River Severn between S2 and the River Teme confluence (Figure 5.2). Of the 11 migrations tracked upstream of T1 by 9 individuals, there were 3 approaches of T2, with the remaining 8 reaching upstream extents between 7 and 13 km downstream of T2 (Figure 5.2). Overall, weirs formed the upstream extent for 64% of migrations tracked upstream from S1a/b, and 41 % of migrations tracked upstream from S2.

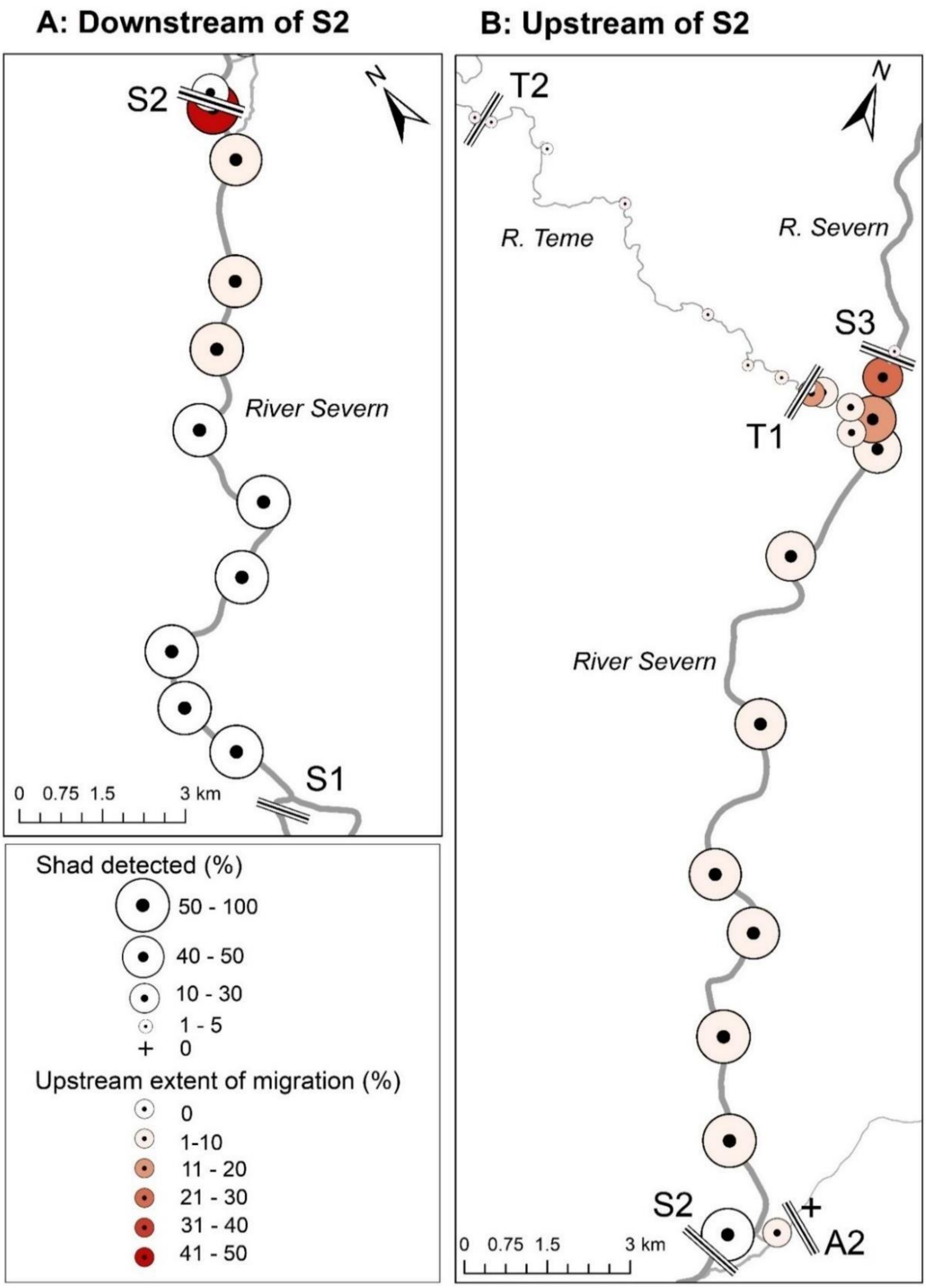

Figure 5.2: The upstream extent of acoustic-tagged twaite shad in the River Severn catchment tracked during spawning migrations in 2018-2020. The percentage of shad reaching each receiver, and the percentage of shad reaching their upstream extent of migration at each receiver, are represented by the size and colour intensity of the circles, respectively. Data is pooled for newly-tagged and returning fish. The weir codes are as in Table 5.1. A: Upstream extent of shad migrations recorded upstream of weir S1 (*n* migrations = 152). B: Upstream extent of shad recorded upstream of weir S2 (*n* migrations = 164).

5.4.3 Individual factors influencing approach of weirs

There were 16 GLMMs that tested the factors influencing approach of S2 and S3/T1 by all fish (Table A8). The best-fitting model retained weir and body length as predictors of weir approach (ΔAIC from null model = 12.5), indicating that shad available to approach S3/T1 were less likely to approach these weirs than those available to approach S2 (Table 5.6, Figure 5.3). While body length was retained in the model and had a positive effect, its effect was non-significant (P = 0.15; Table 5.6, Figure 5.3). There were seven GLMs that tested the likelihood of weir approach by returning fish at S3/T1 (Table A8). The best fitting model (ΔAIC from null model = 1.3) retained the previous approach of S3/T1 as the sole predictor, with the model indicated a marginally significant positive effect of previous approach on approach likelihood (P= 0.06; Table 5.6, Figure 5.3).

Table 5.6: Covariate effects from best-fitting models of weir approach likelihood by twaite shad; a) best fitting generalised linear mixed model including newly tagged and returning fish (Dataset 1); b) best fitting generalised linear model including only returning fish (Dataset 2).

Parameter	**Estimate**	**SE**	**z**	**p**
a)				
(Intercept)	-3.42	2.98	-1.15	0.25
Weir: S3/T1	-	-	-	-
Weir: S2	2.31	0.78	2.97	<0.01
Body length	1.19	0.83	1.43	0.15
b)				
(Intercept)	-0.41	0.65	-0.63	0.53
Previous approach	1.50	0.80	1.88	0.06

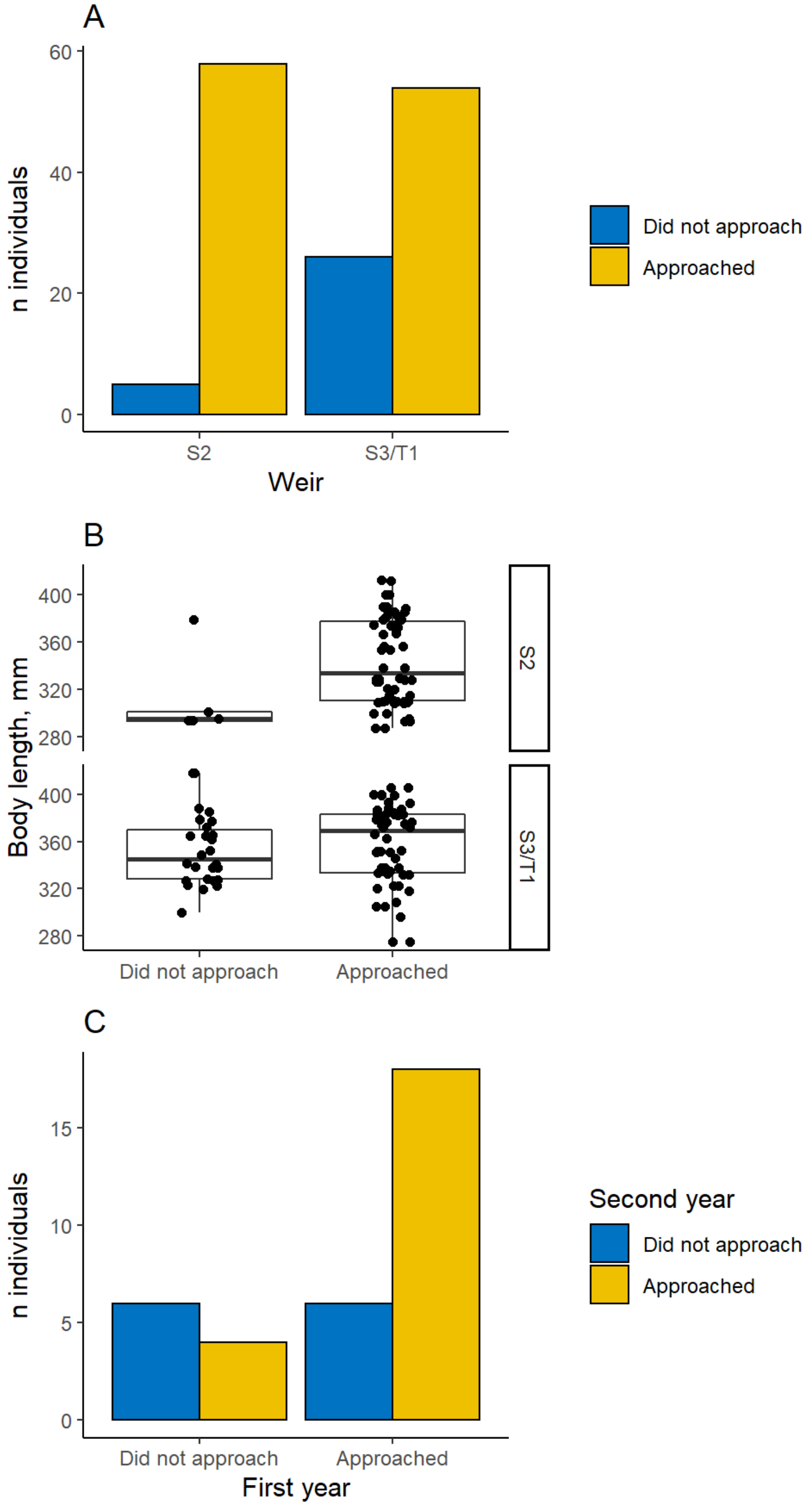

Figure 5.3: Covariates contained within the best-fitting linear mixed models of weir approach likelihood in twaite shad. A: Number of approaching/non-approaching individuals. B: Body length of approaching/non-approaching individuals by weir for newly tagged and returning individuals. C: Number of approaching/non-approaching individuals at weirs S3/T1 by previous approach, for returning individuals.

5.4.4 Individual and environmental factors influencing passage rates of weir S2

There were 32 mixed effects cox models testing the individual and environmental factors influencing passage rates of weir S2 by newly tagged and returning fish (Dataset 1) (Table A9). The best fitting model (ΔAIC from null model = 27.6; Akaike weight = 0.36) revealed that shad passed S2 at a significantly greater rate during higher river level conditions and at higher water temperatures ($p \leq 0.01$; Table 5.7, Figure 5.4). Tagging status had an influence on passage rates, with returning fish passing S2 at a significantly higher rate than newly tagged fish ($P < 0.01$; hazard ratio (HR) = 6.04 (2.11-17.27)), Table 5.7, Figure 5.5). Passage rate was reduced at night compared with daytime, and larger shad had higher passage rates, although these effects were non-significant (Table 5.7).

A further 32 mixed effects cox models tested factors influencing passage rates of weir S2 by returning fish (Dataset 2; Table A10), with the best fitting model (ΔAIC from null model = 19.9; total Akaike weight = 0.23) retained previous success, diel period, river level and water temperature as predictors (Table 5.7, Figure 5.5). Previous passage success significantly increased passage rates for returning fish relative to previously unsuccessful fish ($p = 0.04$; HR = 3.27 (1.07-9.97), Table 5.7, Figure 5.5). Hazard ratios for the other covariates were of the same direction as in Dataset 1, although their magnitude varied (Table 5.7).

Table 5.7: Individual and environmental covariates on passage rate of weir S2 by twaite shad; a) model including newly tagged fish released at weir S1a (Figure 5.1) and returning fish; b) model including only returning fish.

Parameter	**Hazard ratio (95% confidence interval)**	**z**	**p**
a)			
Tagging status: newly tagged	-	-	-
Tagging status: returner	6.04 (2.11-17.27)	3.36	<0.01
River level, m	11.5 (4.43-29.81)	5.02	<0.01
Diel period: Day	-	-	-
Diel period: Night	0.28 (0.06-1.28)	-1.64	0.10
Body length, mm	1.01 (0.99-1.02)	1.20	0.23
Water temperature	1.43 (1.07-1.92)	2.44	0.01
b)			
Previous success: Failed	-	-	-
Previous success: Passed	3.27 (1.07-9.97)	2.08	0.04
River level	21.51 (3.81-121.46)	3.47	<0.01
Diel period:Day	-	-	-
Diel period:Night	0.35 (0.06-1.86)	-1.24	0.22
Water temperature	1.69 (1.2-2.38)	2.98	<0.01

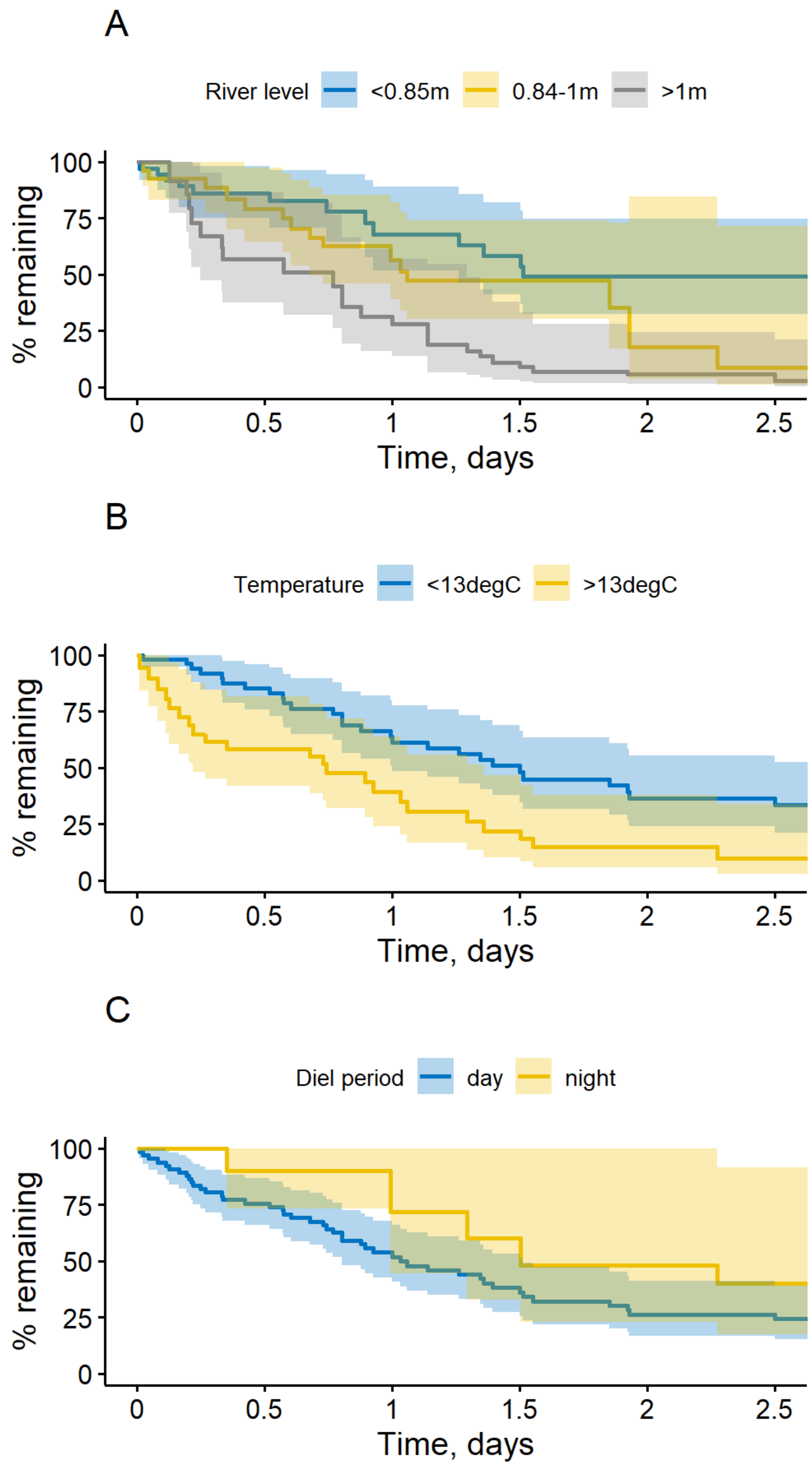

Figure 5.4: Kaplan-Meir survival distributions for passage of weir S2 by acoustic-tagged twaite shad. Covariates effects presented are environmental covariates shown to have a significant effect on passage rates in the top ranked mixed-effects Cox model. Lines represent % of shad that are yet to pass the weir at each time point. For continuous covariates, survival distributions are displayed for representative data categories (Goerig et al. 2020). A: The effect of river level recorded on passage rates. B: The effect of temperature on passage rates. C: The effect of diel period on passage rates.

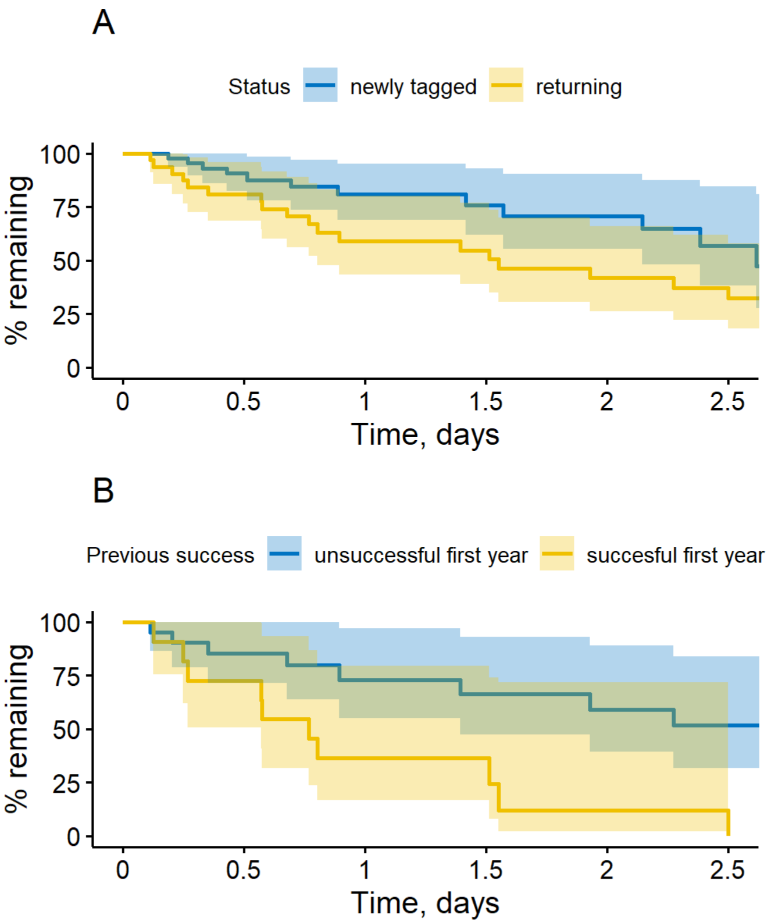

Figure 5.5: Kaplan-Meir survival distributions for passage of weir S2 by acoustic-tagged twaite shad. Covariates effects presented are from individual covariates shown to have a significant effect on passage rates in the top ranked mixed-effects Cox model. Lines represent % of shad that are yet to pass the weir at each time point. For continuous covariates, survival distributions are displayed for representative data categories (Goerig et al. 2020). A: The effect of tagging status (newly-tagged versus returning) on passage rates. B: The effect of previous success on passage rates by returning individuals.

5.5 Discussion

In this study, weirs were substantial barriers to the migration of the threatened anadromous twaite shad. While all weirs in the lower Severn catchment prevented a subset of fish from moving upstream, the weirs did vary in their degree of impact in terms of passage rates and temporal delays to migration. Environmental conditions also influenced passage rates, with episodes of elevated river levels and temperatures important for facilitating passage. Amongst fish returning in their second tracking year, there was evidence for a significant positive effect of previous success on passage rates, potentially suggesting a conserved ability and/or motivation to pass barriers between years. Returning fish also passed at higher rates than newly tagged fish, highlighting the importance of considering tagging effects when assessing barrier impacts using telemetry.

5.5.1 Impact of weirs on shad migration

The proportion of fish that passed each weir was highly variable, but other than the highly tidal weirs initially encountered by the fish in the lower reaches, the proportions that passed were generally lower than the 90 % target recommended for migratory fish at anthropogenic barriers (Lucas & Baras, 2001). This suggests that a substantial percentage of twaite shad were prevented from reaching upstream spawning habitat, a factor which is heavily implicated in the decline of spawning populations of anadromous shads in the River Severn and elsewhere in their range (e.g. Aprahamian et al., 2003a; Limburg & Waldman, 2009; Buffery, 2018). In addition, the weirs imposed migration delays on the fish that were considerable in the context of their overall time in river. Delays to migration have been shown to have negative consequences for reproductive success and survival in anadromous fish generally (Castro-Santos & Letcher, 2010), with delays also potentially subjecting migrants to elevated predation risk (Schmitt et al., 2017; Alcott, Long & Castro-Santos, 2020). Weirs formed the upstream limit of migration for the majority of acoustic-tagged shad, further indicating their restrictive impact on shad distribution in the Severn catchment. Taken together, these findings emphasise the need for passage remediation work in the lower

River Severn catchment, supporting the work that has been continuing on the river in this respect (www.unlockingthesevern.co.uk). Facilitating shad passage at these structures can incorporate barrier removal with the retro-fitting of fish passes that take into account the specific knowledge base on passage requirements for alosines (Haro & Castro-Santos, 2012; Pess et al., 2014; Mulligan et al., 2019). Indeed, the preliminary results here indicated that modifying weir T1 did increase their passage rates, increasing from 0 % pre-modification to 50-67% post-modification, albeit these involved relatively low numbers of tagged individuals.

5.5.2 Factors affecting approach of weirs

While barriers formed the upstream limit of migration for the majority of acoustic-tagged shad in the lower Severn catchment, a subset of individuals within each impounded section did not make approaches to weirs, particularly in the reach of river upstream of S2, and upstream of T1. This finding is potentially indicative of the availability of apparently high-quality spawning habitat in the lower River Teme, which is characterised by shallow, fast-flowing riffle and run habitat generally less than 2m in depth (APEM, 2014; Antognazza et al., 2019). Twaite shad that reached their upstream extent further downstream in the Severn suggest the existence of further spawning sites within deeper waters. There was also evidence that the likelihood of an individual approaching a barrier was repeatable across years, with returning individuals that approached S3 and/or T1 in previous years more likely to approach upon their return. This tentatively suggests these individuals had a conserved motivation to approach and pass barriers, and/or displayed fidelity to areas of previous spawning (Dyer & Brewer, 2020) that were further upstream than non-approaching individuals. This result is relevant to studies of recolonisation following river reconnection, because it suggests there could be a subset of upstream migrants that may be more motivated to exploit newly opened habitat following barrier passage remediation (Pess et al., 2014). However, further work is required to understand the spatial distribution of shad movements within the Severn catchment, including via telemetry studies, and to test whether they do exhibit reach-specific fidelity.

5.5.3 Individual factors affecting weir passage rates

In this study, returning individuals passed a navigation weir at significantly higher rates than newly tagged individuals. This result could indicate a negative effect of capture and/or tagging on barrier passage ability, although a confounding factor in this is that there is a body size increase between tagging one year and its migration the following year. While body length was included in the top-ranked model of passage rates in the first model dataset, its predicted positive effect on passage rates was low, and it was not included in the top-ranked model of passage by returning individuals in the second dataset. Thus, the reduced passage rate by newly tagged individuals was considered to be at least partly due to sublethal tagging effects that affected their migratory behaviour and manifested as their reduced ability and/or motivation to pass weirs in the tagging year. Tagging effects have been a pernicious feature of telemetry studies in alosines (Frank et al., 2009; Eakin, 2017), and this finding supports previous work in alewife *Alosa pseudohaerengus,* which found PIT-tagged returnees had higher passage rates of weirs than newly tagged fish (Nau et al., 2017). Taken together, these results may indicate that returning fish are considered a 'gold-standard' to newly tagged fish for assessing passage at barriers and migration in general. However, their use is clearly only possible in iteroparous species with a relatively high annual survival rate, and the increased costs incurred in generating a reliable sample size of returning individuals using multi-year telemetry may be considerable (Raabe et al., 2019). Further work should thus seek to provide a mechanistic understanding of reduced passage rates in newly tagged fish, which may help mitigate them; possible studies may involve finer-scale telemetry studies and controlled experimental studies to elucidate and separate potential effects of capture, sedation and tagging on key predictors of passage ability such as motivation, orientation, swimming performance (Cooke et al., 2011). This is in addition to further direct comparisons between newly tagged and returning fish on aspects including upstream movement speed and rates of fallback (Frank et al., 2009).

Here, previous success in passage of a navigation weir by twaite shad was also associated with significantly greater passage rates when they returned the following year versus previously unsuccessful fish. This may indicate that learned behaviours enable faster passage of barriers, although a previous study found little evidence for prior experience increasing passage rates in returning flathead catfish *Pylodictis olivaris* and striped bass *Morone saxatilis*, while acknowledging that environmental variability may have masked individual effects (Raabe et al., 2019). Inherent phenotypic (body size, body shape) (Goerig et al., 2020) or behavioural traits may also enable certain individuals to be more successful at passing barriers (Kirk & Caudill, 2017), although there was no evidence for an effect of body size on passage rates amongst returning individuals in this study. A third potential explanation relates to variation in motivation that is driven by spatial fidelity or natal homing. A widely reported feature of shad spawning distributions in fragmented river catchments is that spawning often occurs in areas immediately downstream of weirs (Acolas et al., 2006; López et al., 2007). This was also observed in this study, and might lead to imprinting of juveniles to impounded areas downstream of barriers, resulting in a reduced motivation to progress upstream upon their return. Further, there may also be learned spatial preferences in repeat-spawning adults, whereby they display preferences to using spawning areas that were used in previous years (Pess et al., 2014). Hatchery-reared American shad have demonstrated that imprinting is likely to occur at the tributary level (Hendricks et al., 2002), although the mechanism of imprinting, and precision natal homing and spatial fidelity in alosines is generally poorly understood (Pess et al., 2014).

5.5.4 Environmental factors affecting weir passage rates

Passage of barriers, such as weirs, by fish can be influenced by swimming capacity and attempt rate, which in turn can be influenced by environmental variables, such as water temperature and discharge, as well as barrier characteristics, including head height and the presence of fish passage structures (Castro-Santos, 2004; Bunt, Castro-Santos & Haro, 2012). Here, increasing water temperature had a positive effect on passage rates of weir S2. In upstream-migrating fish, changes in water temperature may invoke physiological and behavioural changes linked to maturation of reproductive organs, factors which in turn increase its motivation to ascend and pass a barrier (Lubejko et al., 2017). Higher temperatures have been shown to reduce the failure rates of alewife *Alosa pseudoharengus* attempting to pass fishways (Franklin et al., 2012), and experimental studies in American shad have shown that temperature can increase attempt rate at velocity barriers, but may reduce swimming endurance, indicating how environmental variability through the migration season may influence barrier passage in alosines (Bayse, McCormick & Castro-Santos, 2019). Other studies have reported increased passage rates within the range of temperatures at which spawning occurs, and attributed this to increased motivation to move upstream and spawn (Raabe et al., 2019).

Increasing river levels downstream of S2 significantly increased passage rates at this weir. Downstream river levels at S2 are affected by both tides and river discharge, and thus the relative effects of discharge and tide on passage are challenging to decouple. Nonetheless, the results suggest that prevailing hydraulic conditions at the weir are an important influence on passage by twaite shad. There are several mechanisms by which hydraulic conditions can influence passage of barriers. Water depth at the entrance to fish passes has been shown to increase passage rates of fish passes in American shad (Mulligan et al., 2019), a finding linked to reduced flow velocities at higher water depths. Passage of alosines may also be negatively affected by noise and entrained air and turbulence, all of which may be influenced by downstream river levels (Haro & Castro-Santos, 2012). Further research is required to understand the influence of temperature and river level on barrier passage rates in twaite shad reported here, potentially incorporating fine scale telemetry studies to assess the impacts of water temperature, turbulence, entrained air and water depth on attempt rate, swimming speed and avoidance behaviours.

Here, there was also some evidence that the passage rates of S2 were greater during the day than at night. Alosines have been shown to prefer daylight hours to migrate upstream (Haro & Castro-Santos, 2012; Raabe et al., 2019), while spawning in twaite shad is highly nocturnal (López et al., 2011). The lower passage rate at night may thus have reflected differences in motivation between day/night approaches, whereby approaches to the weir during the day represented passage attempts and nocturnal approaches represented upstream movements linked to spawning (Acolas et al., 2004; López et al., 2011). In anadromous shads, spawning activity immediately downstream of barriers has been attributed to ‘forced’ spawning of unsuccessful individuals, as well as the presence of relatively high quality habitat immediately downstream of weirs (Acolas et al., 2004; Acolas et al., 2006; López et al., 2011). Further work is thus required to understand potential spatial differences in nocturnal versus diurnal approaches to weirs by shad, with this aiming to improve current understandings of characteristics such as spatial fidelity and motivation.

5.5.5 Summary

This study represents, to the best of our knowledge, the first use of telemetry to assess the impacts of anthropogenic barriers on migration in twaite shad and indicated that weirs can represent substantial impediments to upstream migration in this threatened anadromous fish. While returning individuals to their spawning rivers are a rare feature of barrier passage assessments, their use in this study, enabled by advancements in telemetry technology and tagging protocols, was crucial in their use as ‘controls’ for understanding potential tagging bias and for understanding the effect of previous experience on passage ability. The results nonetheless showed that even with previous experience, migrating fish can still be delayed or unable to pass barriers, with elevated river levels and water temperatures important for passage. Taken together, these results are important contributions to contemporary understandings of anadromous fish migration in fragmented river catchments.