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Can telomere length be estimated from fish scales to inform on individual stress?

Gaëlle Brahy | Aurélie Manicki | Maïka Minjou | François Guéraud | Sylvie Oddou-Muratorio | Jacques Labonne
UMR ECOBIOP 1224, EDENE, Université de Pau et des Pays de l'Adour. 64310, Saint-Pée-sur-Nivelle, France

Telomeres are non coding repetitive sequences of TTAGGG, between 1000 and 30 000 base pairs. They are expectedly shortening with each cell division, individual age and oxidative stress. Relative telomere length (RTL) is hence used as an indicator of stress in many taxa.

Various types of fish tissues have been used to carry out telomere length analysis, but not yet fish scales. Scale sampling is low invasive, cost and storage-effective, and scale collections are widely available databanks. Here we explore the potential for scale collections to provide (1) estimation of RTL, and (2) information on stress.

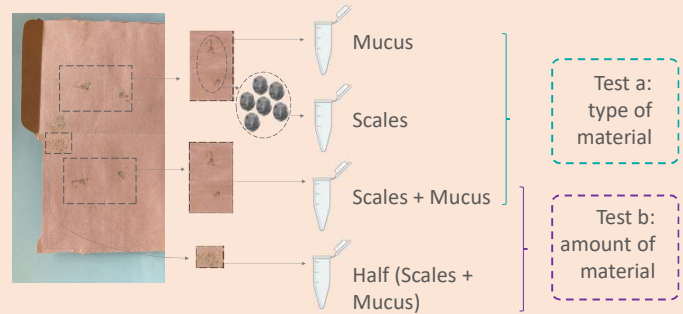
1. Samples and data

INRAE-GISED
Salmo trutta
3 populations
3 age classes
198 individuals

PRELEVEMENT D'ÉCAILLES

ESPECES: DATE: 12/01/2018
LIEU DE CAPTURE: 147
LONGUEUR FOURCHE: 100
SEXE (Mâle ou femelle):
MARQUE: SF145
ANNEE: 2018
AGE:

2. Fish scale sampling: optimization for DNA extraction



3. Individual RTL estimation through qPCR

Principle

2 ng of purified DNA material is used for qPCR

Long telomeres → qPCR Time → Low C_t value

Short telomeres → ++qPCR Time → High C_t value

Threshold fluorescence

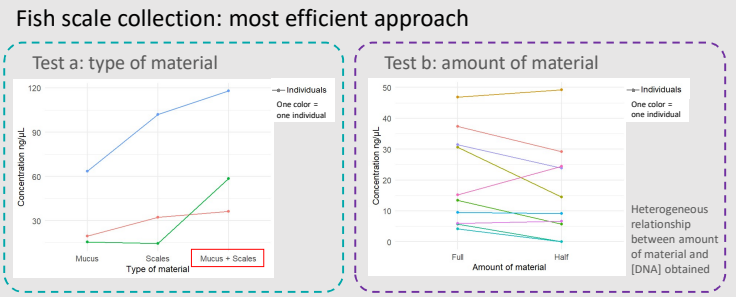
Formula

$$RTL = \frac{E_{telo} \Delta C_{t_{telo}}}{E_{ref} \Delta C_{t_{ref}}}$$

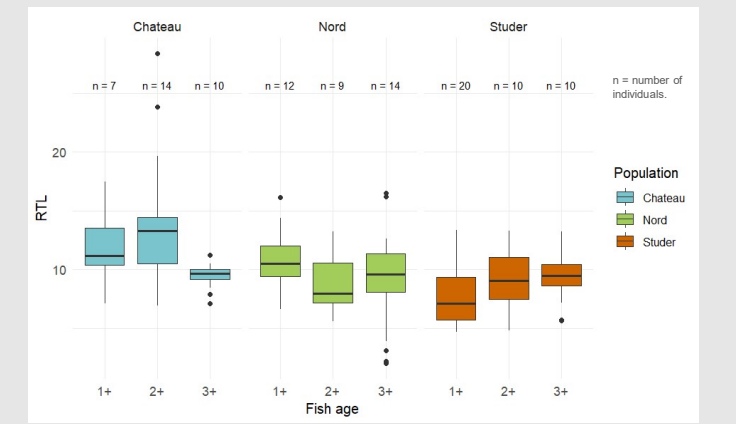
Where $E =$ qPCR efficiency value
 $\Delta C_t = C_{t_{calibrator}} - C_{t_{focal}}$
Ref = reference gene (here, β -actin)

Cawthon, 2002; Naslund et al., 2015

4. Results



RTL in 3 populations, 3 age classes



2 replicates per individual
106 (53%) individuals with a C_t difference between replicates < 0,5
165 (83%) individuals with a C_t difference between replicates < 1

- ## 5. Take home message
- Greater DNA concentrations are obtained when both **mucus and scales** are used
 - Amount of available material does not limit DNA extraction
 - **RTL can be estimated through qPCR from scale material**
 - Intra-individual variability in RTL estimation calls for tailored statistical approach
 - Our results open a new opportunity to **valorise scale collections**
 - There is a **potential to understand RTL variation with stress**

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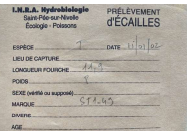
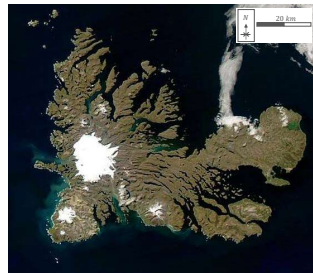
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1. Samples and data



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4. Individual RTL estimation through qPCR: formula

$$RTL = \frac{E_{telo} \Delta C_{t_{telo}}}{E_{ref} \Delta C_{t_{ref}}}$$

Where

E = qPCR efficiency value

$\Delta C_t = C_{t_{calibrator}} - C_{t_{focal}}$

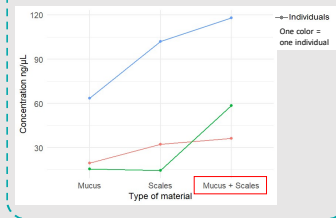
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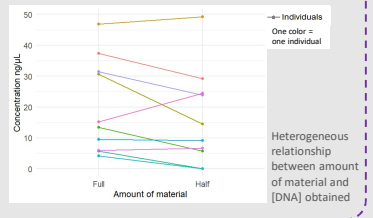
5. Results

Fish scale collection: most efficient approach

Test a: type of material

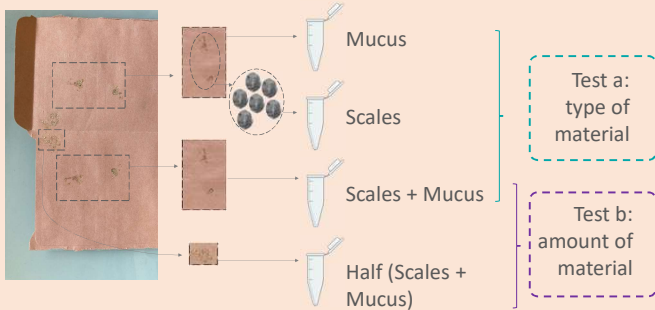


Test b: amount of material



Heterogeneous relationship between amount of material and [DNA] obtained

2. Fish scale sampling: optimization for DNA extraction

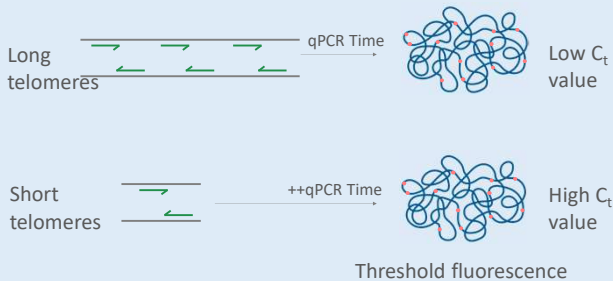


Test a: type of material

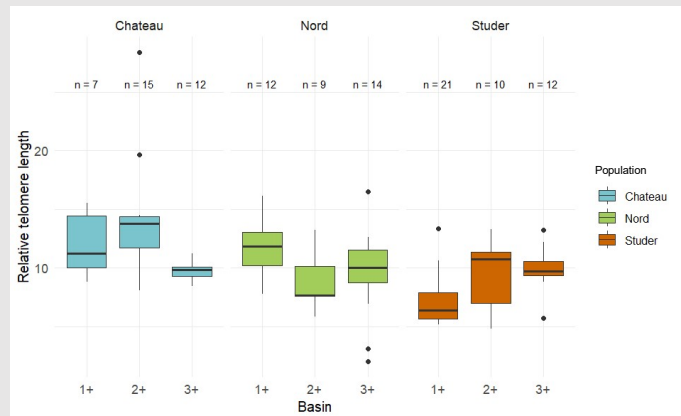
Test b: amount of material

3. Individual RTL estimation through qPCR: principle

2 ng of purified DNA material is used for qPCR



RTL in 3 populations, 3 age classes



2 replicates per individual

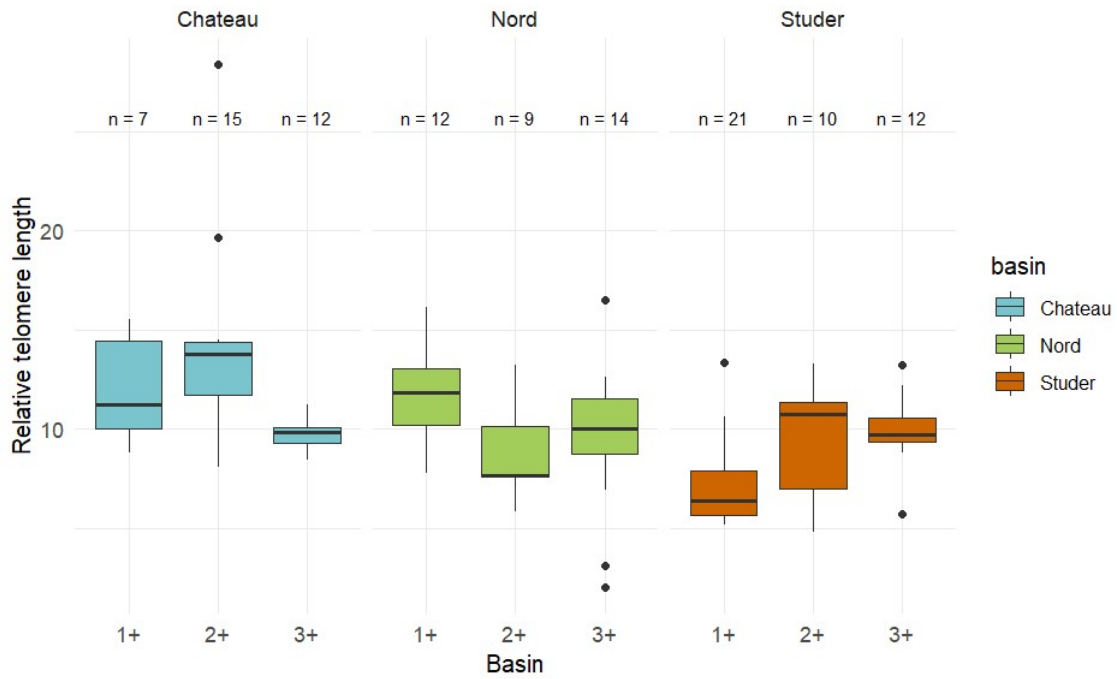
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diff entre réplcats <0.5



diff entre réplcats <1

