**SUPPORTING INFORMATION**

**Evolutionary drivers of diversification and distribution of a southern temperate stream fish assemblage: testing the role of historical isolation and spatial range expansion**

Albert Chakona, Ernst R. Swartz and Gavin Gouws

**Appendix S1** **Sampling localities in the south-western Cape Floristic Region of South Africa.** Tributaries sampled for *Galaxias* (*Gal*), *Pseudobarbus* (*Pse*) and *Sandelia* (*San*) from the south-western CFR. Locality codes and geographic coordinates are given. The number of individuals that were sequenced per locality is indicated. Blank space indicates that individuals of that genus were not collected at that locality.

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | Sample Code | River | System | Latitude | Longitude | *Gal* | *Pse* | *San* |
| 1 | AC08A07 | Vink | Breede | -33.73422222 | 19.82030556 |  | 2 |  |
| 2 | AC08A10 | Vink | Breede | -33.73022222 | 19.82355556 | 2 |  |  |
| 3 | AC08A11 | Noree | Breede | -33.72494444 | 19.81547222 | 1 |  |  |
| 4 | AC08A12 | Hoeks | Breede | -34.02316667 | 19.83852778 |  |  | 1 |
| 5 | AC08A13 | Hoeks | Breede | -34.01091667 | 19.83922222 | 1 |  | 1 |
| 6 | AC08A14 | Hoeks | Breede | -34.00963889 | 19.83836111 | 1 | 2 |  |
| 7 | AC08A17 | Noree | Breede | -33.73277778 | 19.81361111 |  | 2 |  |
| 8 | AC08A18 | Hex | Breede | -33.49877778 | 19.53061111 | 2 |  |  |
| 9 | AC08A19 | Hex | Breede | -33.52894444 | 19.54008333 | 3 | 2 | 2 |
| 10 | AC08A20 | Amandel | Breede | -33.52402778 | 19.54266667 | 2 | 2 |  |
| 11 | AC08A21 | Amandel | Breede | -33.50661111 | 19.49497222 |  |  | 1 |
| 12 | AC08A31 | Amandel | Breede | -33.99380556 | 19.18263889 |  | 2 |  |
| 13 | AC08A32 | Amandel | Breede | -33.98900000 | 19.18402778 | 2 |  | 2 |
| 14 | AC08A38 | Amandel | Breede | -33.97447222 | 19.19175000 | 3 | 2 |  |
| 15 | AC08A42 | Amandel | Breede | -33.98297222 | 19.18963889 | 2 |  |  |
| 16 | AC08A52 | Wit | Breede | -33.57322222 | 19.13797222 |  | 2 | 2 |
| 17 | AC09A08 | Tradou | Breede | -33.95677778 | 20.70788889 |  | 1 |  |
| 18 | AC09A09 | Tradou | Breede | -33.94155556 | 20.70733333 |  | 2 |  |
| 19 | AC09A42 | Wolwekloof | Breede | -33.56413889 | 19.12933333 | 4 |  |  |
| 20 | AC09A43 | Wolwekloof | Breede | -33.56505556 | 19.13144444 |  | 2 | 2 |
| 21 | AC09A46 | Titus | Breede | -33.40533333 | 19.41805556 |  | 2 | 2 |
| 22 | AC09A50 | Keissies | Breede | -33.71050000 | 19.93105556 | 2 |  |  |
| 23 | AC09A51 | Keissies | Breede | -33.70352778 | 19.91086111 | 3 |  |  |
| 24 | AC09A54 | Keissies | Breede | -33.71050000 | 19.93105556 | 1 |  |  |
| 25 | AC09A58 | Koo | Breede | -33.59450000 | 19.75811111 |  | 2 | 2 |
| 26 | AC09A61 | Die Brak | Breede | -33.57644444 | 19.75163889 |  | 2 |  |
| 27 | AC09A63 | Elandskloof | Breede | -34.03055556 | 19.39000000 |  | 2 |  |
| 28 | AC09A64 | Elandskloof | Breede | -34.03461111 | 19.39327778 |  | 2 |  |
| 29 | AC09A65 | Baviaans | Breede | -34.02705556 | 19.55588889 |  | 2 | 2 |
| 30 | AC09A70 | Gobos | Breede | -34.03661111 | 19.63613889 | 2 | 2 | 2 |
| 31 | AC09A71 | Gobos | Breede | -34.03833333 | 19.62844444 | 1 |  |  |
| 32 | AC09A72 | Gobos | Breede | -34.03638889 | 19.62575000 | 1 | 2 |  |
| 33 | AC09A73 | Ganskraal | Breede | -34.11577778 | 19.81394444 |  | 2 | 1 |
| 34 | AC09A74 | Happy Valley | Breede | -34.11183333 | 19.81530556 | 2 | 2 |  |
| 35 | AC09A75 | Happy Valley | Breede | -34.11222222 | 19.69797222 | 2 |  | 1 |
| 36 | AC09A76 | Ganskraal | Breede | -34.12547222 | 19.81147222 | 2 |  |  |
| 37 | AC09A78 | Boks | Breede | -34.07963889 | 19.82952778 | 4 |  |  |
| 38 | AC09A79 | Boks | Breede | -34.09602778 | 19.82841667 |  | 2 |  |
| 39 | AC09B14 | Boesmans | Breede | -34.01113889 | 20.00722222 |  | 1 |  |
| 40 | AC09B15 | Boesmans | Breede | -34.05100000 | 19.95300000 |  |  | 2 |

Appendix S1: continued...

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|  | Sample Code | River | System | Latitude | Longitude | *Gal* | *Pse* | *San* |
| 41 | AC09B17 | Boesmans | Breede | -34.04158333 | 19.96205556 |  | 1 |  |
| 42 | AC09B18 | Poesjenels | Breede | -33.96547222 | 19.58336111 |  |  | 2 |
| 43 | AC09B20 | Poesjenels | Breede | -33.97916667 | 19.55761111 | 1 |  |  |
| 44 | AC09B22 | Dorn | Breede | -33.85333333 | 19.47580556 |  |  | 1 |
| 45 | AC09B23 | Riesvlei | Breede | -33.89316667 | 19.44344444 |  |  | 1 |
| 46 | AC09B25 | Dorn | Breede | -33.93294444 | 19.46161111 | 1 |  |  |
| 47 | AC09B27 | Stettyn | Breede | -33.86977778 | 19.34750000 | 1 |  |  |
| 48 | AC09B30 | Stettyn | Breede | -33.86847222 | 19.34725000 |  | 2 |  |
| 49 | AC09B33 | Holsloot | Breede | -33.82883333 | 19.27161111 | 2 |  | 2 |
| 50 | AC09B35 | Agter-Vink | Breede | -33.67155556 | 19.74255556 | 2 |  |  |
| 51 | AC09B38 | Spreeudrifspruit | Breede | -33.98316667 | 19.32925000 |  | 2 | 2 |
| 52 | AC09B39 | Bereaville | Breede | -34.05930556 | 19.47736111 |  |  | 2 |
| 53 | AC09B41 | Bereaville | Breede | -34.02705556 | 19.48236111 | 5 |  |  |
| 54 | AC09B42 | Bereaville | Breede | -34.03036111 | 19.48377778 |  | 2 |  |
| 55 | AC09B43 | Riviersonderend | Breede | -34.06227778 | 19.07013889 |  | 2 |  |
| 56 | AC09B44 | Riviersonderend | Breede | -34.06741667 | 19.07566667 | 4 |  |  |
| 57 | AC09B46 |  | Breede | -34.05486111 | 19.09744444 | 2 |  |  |
| 58 | AC09C25 |   | Breede | -34.37052778 | 20.64566667 |  | 4 | 1 |
| 59 | AC09C26 |   | Breede | -34.38255556 | 20.67505556 |  |  | 1 |
| 60 | AC09C27 |   | Breede | -34.38694444 | 20.68622222 |  |  | 1 |
| 61 | AC09C56 |   | Breede | -34.04236111 | 19.53716667 | 2 |  |  |
| 62 | AC09C57 |   | Breede | -34.07666667 | 19.64325000 | 2 |  |  |
| 63 | AC09C59 | Gobos | Breede | -34.05344444 | 19.61366667 | 2 |  |  |
| 64 | AC09C61 |  | Breede | -34.06538889 | 19.65883333 | 2 |  | 2 |
| 65 | AC09C63 | Keurbooms | Breede | -34.00441667 | 20.37713889 | 4 |  |  |
| 66 | AC09C65 | Soetmelks | Breede | -34.11238889 | 19.74552778 | 2 |  |  |
| 67 | AC09C66 | Soetmelksvlei | Breede | -34.10330556 | 19.74122222 |  | 2 |  |
| 68 | AC09C67 | Krom | Breede | -34.11166667 | 19.79566667 | 1 | 2 |  |
| 69 | AC09C69 | Klip | Breede | -34.02463889 | 20.41805556 | 2 | 2 | 2 |
| 70 | AC09C71 | Leeu  | Breede | -33.97855556 | 20.34202778 | 1 |  |  |
| 71 | AC09C72 | Leeu | Breede | -33.99508333 | 20.33719444 |  | 2 |  |
| 72 | AC09C74 | Keurbooms | Breede | -33.98561111 | 20.37300000 |  | 3 |  |
| 73 | AC09C75 | Keurbooms | Breede | -33.99644444 | 20.37375000 | 2 |  |  |
| 74 | AC09C77 | Koornings | Breede | -34.01230556 | 20.45458333 | 2 | 1 | 2 |
| 75 | AC09C79 | Hartbees | Breede | -34.01230556 | 20.45458333 | 3 | 1 |  |
| 76 | AC09C81 | Bothaspruit | Breede | -33.56247222 | 19.34075000 | 2 | 5 | 1 |
| 77 | AC09C82 | Du Toit | Breede | -33.96702778 | 19.16566667 | 4 |  | 2 |
| 78 | AC09C84 | Slanghoek | Breede | -33.61155556 | 19.22402778 | 2 |  |  |
| 79 | AC09C85 | Slanghoek | Breede | -33.65522222 | 19.21797222 |  |  | 1 |
| 80 | AC09D08 | Breede | Breede | -33.68705556 | 19.42683333 | 4 |  |  |

Appendix S1: continued…

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | Sample Code | River | System | Latitude | Longitude | *Gal* | *Pse* | *San* |
| 81 | AC09D09 | Breede | Breede | -33.52002778 | 19.18552778 | 2 |  |  |
| 82 | AC09D16 | Du Toit | Breede | -33.93722222 | 19.16800000 |  | 4 |  |
| 83 | AC09D18 | Bothaspruit | Breede | -33.59952778 | 19.32330556 |  | 1 |  |
| 84 | AC09D20 | Wabooms | Breede | -33.50252778 | 19.27063889 | 2 |  |  |
| 85 | AC09D23 | Koekedou | Breede | -33.35911111 | 19.29547222 |  |  | 2 |
| 86 | AC09D25 | Wit tributary | Breede | -33.53744444 | 19.14302778 |  | 2 |  |
| 87 | AC09D28 | Krom | Breede | -33.72277778 | 19.11294444 |  | 6 |  |
| 88 | AC09D32 | Smallblaar | Breede | -33.69050000 | 19.31622222 | 2 |  |  |
| 89 | AC09D33 | Slanghoek | Breede | -33.66619444 | 19.24638889 | 2 |  |  |
| 90 | AC09D34 | Riviersonderend | Breede | -34.06230556 | 19.07050000 |  | 8 |  |
| 91 | AC09D39 | Sandrifs Dam | Breede | -33.43613889 | 19.57088889 |  |  | 2 |
| 92 | AC09D41 | Buffeljags  | Breede | -33.99916667 | 20.60455556 | 2 |  |  |
| 93 | ES07A21 | Baviaans | Breede | -34.02705556 | 19.55588889 | 2 |  |  |
| 94 | gz32 | Happy Valley | Breede | -34.09650000 | 19.73605556 | 1 |  |  |
| 95 | gz33 | Elandskloof | Breede | -34.05202778 | 19.41627778 | 1 |  |  |
| 96 | AC09B13 | Duiwenhoks | Duiwenhoks | -33.99766667 | 21.10583333 |  |  | 2 |
| 97 | AC09C32 | Duiwenhoks | Duiwenhoks | -33.99480556 | 21.07519444 | 2 |  |  |
| 98 | AC09C33 | Duiwenhoks | Duiwenhoks | -33.97819444 | 21.03244444 | 2 |  | 2 |
| 99 | AC09C35 | Duiwenhoks | Duiwenhoks | -33.97041667 | 20.98783333 | 2 |  | 2 |
| 100 | AC09C36 | Duiwenhoks | Duiwenhoks | -33.99211111 | 20.85430556 | 1 |  | 2 |
| 101 | AC09C38 | Duiwenhoks | Duiwenhoks | -34.00705556 | 20.88383333 |  | 8 |  |
| 102 | AC09C39 | Duiwenhoks  | Duiwenhoks | -34.01963889 | 20.93455556 |  | 4 | 2 |
| 103 | AC08A58 | Vette | Goukou | -34.02125000 | 21.22500000 | 4 | 2 | 2 |
| 104 | AC09B01 | Goukou | Goukou | -34.00036111 | 21.41800000 | 7 |  | 2 |
| 105 | AC09B04 | Kleinkruis | Goukou | -34.01719444 | 21.29058333 | 2 | 2 |  |
| 106 | AC09B06 | Kleinkruis | Goukou | -33.99900000 | 21.29138889 |  |  | 2 |
| 107 | AC09B07 | Kleinkruis | Goukou | -34.00272222 | 21.29222222 |  | 4 |  |
| 108 | AC09B10 | Kleinkruis | Goukou | -33.96108333 | 21.30677778 |  |  | 2 |
| 109 | AC09B11 | Vette | Goukou | -33.94430556 | 21.25419444 |  |  | 2 |
| 110 | AC09B12 | Vette | Goukou | -33.98491667 | 21.20716667 |  | 8 |  |
| 111 | AC09C31 | Korinte | Goukou | -33.98883333 | 21.15794444 | 2 |  | 2 |
| 112 | AC09A28 | Haelkraal | Haelkraal | -34.66763889 | 19.57527778 | 3 |  |  |
| 113 | AC09A29 | Haelkraal | Haelkraal | -34.66775000 | 19.57791667 |  |  | 2 |
| 114 | AC09A30 | Haelkraal | Haelkraal | -34.67683333 | 19.58697222 | 6 |  |  |
| 115 | ES07A38 | Haelkraal | Haelkraal | -34.67083333 | 19.54300000 | 2 |  |  |
| 116 | AC08A22 | Kars | Heuningnes | -34.41316667 | 19.82041667 | 2 | 2 |  |
| 117 | AC08A23 | Kars | Heuningnes | -34.41330556 | 19.82080556 |  |  | 4 |
| 118 | AC08A24 | Kars | Heuningnes | -34.36372222 | 19.78255556 |  | 2 | 1 |
| 119 | AC08A25 | Kars | Heuningnes | -34.36855556 | 19.78308333 |  |  | 1 |
| 120 | AC09A23 | Grashoek | Heuningnes | -34.58238889 | 19.96841667 |  |  | 2 |

Appendix S1: continued…

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | Sample Code | River | System | Latitude | Longitude | *Gal* | *Pse* | *San* |
| 121 | AC09A24 | Grashoek | Heuningnes | -34.57400000 | 19.94888889 | 1 | 3 |  |
| 122 | AC09A25 | Nuwejaars | Heuningnes | -34.55497222 | 19.88583333 | 1 |  |  |
| 123 | AC09A26 | Nuwejaars | Heuningnes | -34.54230556 | 19.81916667 | 2 |  | 2 |
| 124 | AC09A27 | Nuwejaars | Heuningnes | -34.57836111 | 19.75758333 | 3 | 4 | 2 |
| 125 | AC09B51 | Van Zyl farm tributary | Heuningnes | -34.51836111 | 19.97591667 | 1 |  |  |
| 126 | AC09C12 | Koue | Heuningnes | -34.56136111 | 19.63094444 | 2 |  |  |
| 127 | AC09C15 | Kars | Heuningnes | -34.46783333 | 19.82311111 | 2 |  | 1 |
| 128 | AC09C16 | Kars | Heuningnes | -34.45166667 | 19.90116667 | 3 |  | 2 |
| 129 | AC09C17 | Kars | Heuningnes | -34.41677778 | 19.80119444 |  | 1 |  |
| 130 | AC09B50 | Klein | Klein | -34.27619444 | 19.52513889 |  |  | 2 |
| 131 | AC09C05 | Hartbees | Klein | -34.32511111 | 19.37447222 | 4 |  |  |
| 132 | AC09C07 | Hartbees | Klein | -34.33419444 | 19.35850000 | 4 |  | 1 |
| 133 | ES07A42 | Onrus | Onrus | -34.37825000 | 19.24000000 | 4 |  |  |
| 134 | AC09B49 | Palmiet | Palmiet | -34.06919444 | 19.05072222 |  |  | 2 |
| 135 | AC09B55 | Palmiet | Palmiet | -34.09308333 | 19.05136111 | 2 |  |  |
| 136 | AC09C18 | Ratel | Ratel | -34.70725000 | 19.69733333 | 2 |  |  |
| 137 | AC09C19 | Ratel | Ratel | -34.67011111 | 19.69138889 | 4 |  |  |
| 138 | ES07A36 | Ratel | Ratel | -34.75330556 | 19.73047222 | 4 |  |  |
| 139 | AC09A32 |  | Uilkraals | -34.56222222 | 19.45513889 | 2 |  |  |
| 140 | AC09A33 | Uilkraals | Uilkraals | -34.57366667 | 19.47944444 | 4 |  | 1 |
| 141 | AC09A34 | Boesmans | Uilkraals | -34.59836111 | 19.59222222 | 4 |  |  |
| 142 | AC09A36 | Uilkraals | Uilkraals | -34.54366667 | 19.51519444 |  |  | 1 |
| 143 | AC09A37 | Uilkraals | Uilkraals | -34.52697222 | 19.53927778 | 6 |  |  |
| 144 | AC09A38 | Uilkraals | Uilkraals | -34.46708333 | 19.59711111 |  |  | 1 |
| 145 | AC09C11 | Uilkraals | Uilkraals | -34.51363889 | 19.61202778 | 2 |  |  |
| 146 | GZ42 | Slang | Uilkraals | -34.51338889 | 19.61216667 | 1 |  |  |
| Outgroups | *G*. ‘mollis’ | Leeu | Berg | -33.15591667 | 19.05058333 |  |  |  |
|  | *S*. ‘berg’ | Leeu | Berg | -33.15591667 | 19.05058333 |  |  |  |
|  | *P. asper* | Olifants | Gouritz | -33.62150000 | 21.94819444 |  |  |  |
|  | *P. tenuis* | Olifants | Gouritz | -33.62150000 | 21.94819444 |  |  |  |