MOLECULAR ECOLOGY RESOURCES

Supplemental Information for:

"A chromosome-level genome assembly enables the identification of the follicule stimulating hormone receptor as the master sex determining gene in the flatfish *Solea senegalensis*"

de la Herrán R, Hermida M, Rubiolo J, Gómez-Garrido J, Cruz F, Robles F, Navajas-Pérez R, Blanco, A, Villamayor PR, Torres D, Sánchez-Quinteiro P, Ramirez D, Rodríguez ME, Arias-Pérez A, Cross I, Duncan N, Martínez-Peña T, Riaza A, Millán A, De Rosa MC, Pirolli D, Gut M, Bouza C, Robledo D, Rebordinos L, Alioto T, Ruíz-Rejón C, Martínez P

Table of Contents:

MOLECULAR ECOLOGY
RESOURCES

1. SUPPLEMENTARY TABLES

Table S1: Genome assembly statistics of *Solea senegalensis*; fSolSen1_LG: set of contigs anchored to linkage groups (LG) using the genetic map.

Assembly	*fSolSen1_LG*	*fSolSen1.LG_plus_unanchored contigs*
Scaffold N50	29.025.067	28.642.601
Scaffold L50	9	10
Contig N50	23.439.045	23.439.045
Contig L50	12	12
Longest contig (Mb)	30.1	30.1
Total Length (bp)	606.883.213	613.860.701
No. Contigs	51	82
BUSCO v 5.4.0*	C:98.4%[S:97.4%,D:1.0%],F:0.3%,M:1.3%,n:3640	C:98.4%[S:97.4%,D:1.0%],F:0.3%,M:1.3%,n:3640
QV	44,1253	43,1767
Kmer completeness	98,0442	98,1823

* using the actinopterygii_odb10 database

MOLECULAR ECOLOGY RESOURCES

Table S2. Statistics of TE-derived sequence and other simple repeats in the genome of *Solea senegalensis*.

	number of elements	length (bp)	% genome sequence
Retroelements	**69002**	**13535052**	**2.23**
SINEs:	15162	1329007	0.22
Penelope	2164	138108	0.02
LINEs:	31457	7794235	1.28
CRE/SLACS	0	0	0.00
L2/CR1/Rex	20727	4996872	0.82
R1/LOA/Jockey	1023	213578	0.04
R2/R4/NeSL	454	171826	0.03
RTE/Bov-B	1620	614510	0.10
L1/CIN4	4351	1307527	0.22
LTR:	22383	4411810	0.73
BEL/Pao	1456	776782	0.13
Ty1/Copia	46	49522	0.01
Gypsy/DIRS1	8429	2361194	0.39
Retroviral	5460	453444	0.07
DNA transposons	**158069**	**15032076**	**2.48**
hobo-Activator	63226	5491718	0.90
Tc1-IS630-Pogo	14261	2782338	0.46
En-Spm	0	0	0.00
MuDR-IS905	0	0	0.00
PiggyBac	776	82949	0.01
Tourist/Harbinger	5608	490156	0.08
Other (Mirage, P-element, Transib)	759	38789	0.01
Unclassified:	**881**	**151676**	**0.02**
Total interspersed repeats		**28718804**	**4.73**
Rolling-circles	**8594**	**1496522**	**0.25**
Small RNA:	**4066**	**320172**	**0.05**
Satellites:	**1711**	**273438**	**0.05**
Simple repeats:	**401374**	**17082324**	**2.81**
Low complexity:	**38393**	**1952345**	**0.32**

Note. N.A.- Not available. Superfamilies contributing < 1 kb of the genome were not included.

MOLECULAR ECOLOGY
RESOURCES

Table S3: Genome annotation statistics of protein coding genes in the genome of *Solea senegalensis*

	fSolSen1A annotation
Number of protein-coding genes	24,264
Median gene length (bp)	7,566
Number of transcripts	40,511
Number of exons	277,235
Number of coding exons	263,350
Median UTR length (bp)	950
Median intron length (bp)	389
Exons/transcript	12.79
Transcripts/gene	1.67
Multi-exonic transcripts	95.47%
Gene density (gene / Mb)	39.53

MOLECULAR ECOLOGY
RESOURCES

Table S4: Absolute number of reads and averages obtained by the 2b-RAD-seq method for SNP genotyping in three families (F1, F2, F3) of *Solea senegalensis* and their offspring across the different filtering steps of the established pipeline from the initial raw reads to the final valid alignment against the genome for constructing a highly dense genetic map.

Sample	Initial Raw Reads	Length Filter	Quality Filter	Enzyme Filter	Failed to align	Multiple alignments	Valid alignment
Total	1029218660	1001426201	993835057	853403269	21986669	51779704	779636896
Average	4379654	4261388	4229085	3631503	93560	220339	3317604
Average parents	11970770	11879489	11799911	10360318	252464	646145	9461709
Average offspring	4180760	4061787	4030723	3455202	89397	209183	3156623
Female F1	14002206	13888873	13810304	11982174	281479	762680	10938015
Female F2	12312486	12226228	12111336	10621377	329428	639317	9652632
Female F3	9997256	9923181	9852344	8662808	225957	542630	7894221
Male F1	10016350	9938872	9888228	8569591	178864	536695	7854032
Male F2	12808229	12709151	12639846	11351330	238227	717153	10395950
Male F3	12688090	12590629	12497406	10974628	260830	678396	10035402
F1-006	4413840	4344937	4307867	3768961	87274	233929	3447758
F1-008	3666714	3611929	3495710	3146501	140701	182410	2823390
F1-009	5027743	4920968	4889721	4266791	91809	265114	3909868
F1-019	3852267	3671241	3647833	2653823	63447	162690	2427686
F1-021	4390580	4337712	4300555	3844703	86421	231128	3527154
F1-022	3910252	3846840	3820691	3330801	68210	203225	3059366
F1-023	3893544	3832982	3799578	3402484	78516	206957	3117011
F1-025	3182528	3140974	3118902	2781342	76816	170946	2533580
F1-026	2972834	2924260	2904233	2549665	47002	155183	2347480
F1-027	3903100	3858641	3819399	3452926	86438	207286	3159202

F1-028	4174401	4115609	4089034	3628407	72986	217587	3337834
F1-029	5229229	5151337	5102856	4573048	101605	276089	4195354
F1-031	5565014	5497289	5459441	4920586	90141	299745	4530700
F1-032	4611506	4547406	4511559	4068347	80389	257526	3730432
F1-034	3261569	3194176	3173621	2727962	90157	165026	2472779
F1-035	2921248	2887193	2867603	2563010	49419	158127	2355464
F1-039	1812900	1794940	1783069	1661965	32877	102150	1526938
F1-040	4284851	4156093	4119996	3443413	80337	212865	3150211
F1-041	4095509	4011103	3982988	3462866	73521	210987	3178358
F1-042	4646599	4529026	4497091	3956385	73487	239699	3643199
F1-045	5265351	5191229	5141690	4702032	98705	291243	4312084
F1-048	5324940	5267752	5190548	4731125	128743	285886	4316496
F1-049	4223264	4174778	4147382	3800597	68575	230072	3501950
F1-051	3529207	3491100	3467129	2995175	56945	180898	2757332
F1-053	4032225	3994199	3966569	3655023	68730	218808	3367485
F1-054	3955994	3894433	3862495	3448692	66954	207040	3174698
F1-056	5445428	5373933	5336167	4856151	99846	289968	4466337
F1-057	3971600	3925590	3890334	3544836	73467	217464	3253905
F1-058	3770842	3733574	3704860	3396012	66200	207344	3122468
F1-059	3749448	3657639	3628320	3189013	66063	192405	2930545
F1-060	3137886	3108240	3083444	2822729	57047	169553	2596129
F1-061	2808314	2777364	2758693	2513838	49848	153201	2310789
F1-062	4017941	3978012	3940662	3585872	77217	216313	3292342
F1-063	2998274	2925859	2894590	2501681	63371	150653	2287657
F1-064	4201494	4148311	4110971	3781068	78359	225659	3477050

F1-065	4093130	4045976	4015993	3683689	77762	225360	3380567
F1-066	2521603	2491865	2471041	2230273	55181	134747	2040345
F1-067	5598446	5522204	5474858	4925227	101737	297449	4526041
F1-068	5073112	5026860	4990137	4581770	93371	277089	4211310
F1-070	4829496	4783506	4739173	4319234	91630	261340	3966264
F1-071	5051962	4992014	4948775	4492966	94052	266155	4132759
F1-072	4982384	4907681	4867893	4416645	96884	256895	4062866
F1-073	5431754	5357408	5320234	4808602	103727	288662	4416213
F1-074	3678193	3632496	3596041	3254967	84751	194223	2975993
F1-075	2773797	2736147	2716141	2400688	52068	144012	2204608
F1-076	3683515	3537941	3512895	2949541	59250	172129	2718162
F1-077	4549726	4505057	4471062	4109162	89279	246025	3773858
F1-078	3843200	3801752	3752602	3427126	94266	201628	3131232
F1-081	3504652	3446281	3398707	3000508	81876	180055	2738577
F1-082	4199058	4098416	4038650	3556573	101362	206674	3248537
F1-083	3800089	3732767	3706756	3232657	73333	193991	2965333
F1-084	4443881	4365889	4310242	3810841	105326	231753	3473762
F1-085	3049383	2863489	2837342	2240799	51802	138764	2050233
F1-086	4688584	4611590	4582128	3965648	91414	240765	3633469
F1-087	4811407	4740947	4707563	4225447	88580	257206	3879661
F1-089	3729402	3533364	3498587	2902474	67629	178039	2656806
F1-090	5652011	5535699	5474204	4861098	120478	288390	4452230
F1-091	5368231	4960155	4922951	4044541	85811	244342	3714388
F1-092	4411887	4334727	4306304	3820008	76855	232378	3510775
F1-093	4590407	4462764	4433409	3762747	78549	227004	3457194

F1-095	4187487	4019690	3968401	3350830	82169	205148	3063513
F1-096	2964780	2928235	2907442	2625200	45786	159049	2420365
F1-097	4501444	4460550	4393848	4081650	103846	240157	3737647
F1-098	6420470	6337101	6285420	5731657	125659	344119	5261879
F1-099	4118611	4029172	3986262	3552101	89126	217354	3245621
F1-100	4794174	4728529	4691730	4266486	82250	258133	3926103
F1-101	5361893	5195127	5146521	4509167	102634	273547	4132986
F1-102	4592528	4524720	4484230	3986673	80992	241518	3664163
F1-103	3448938	3405464	3382970	3058775	60484	187927	2810364
F1-104	2660592	2624015	2607392	2347952	42116	144104	2161732
F1-105	5270220	5195206	5149353	4581534	103995	278111	4199428
F1-106	2710036	2648786	2632257	2327757	44042	143367	2140348
F1-107	5342226	5281203	5236505	4773132	140686	278973	4353473
F1-108	3789296	3664297	3627729	3113200	77564	183871	2851765
F1-109	4225729	3970239	3943453	3123793	57692	192975	2873126
F1-110	3774004	3556984	3533513	2924629	57374	179520	2687735
F1-111	4021370	3966685	3928900	3506233	84435	212269	3209529
F1-112	3348119	3284145	3263642	2887845	49898	175800	2662147
F1-113	3288104	3248914	3219279	2925648	60379	177174	2688095
F1-114	4443415	4187803	4153652	3536005	81554	214160	3240291
F1-115	3530183	3479954	3456705	3128574	66297	188307	2873970
F2-001	4301116	4208103	4176953	3576795	80792	211668	3284335
F2-002	3999245	3896745	3876642	3240971	68140	189676	2983155
F2-006	4274236	4146508	4125165	3408210	67061	208445	3132704
F2-008	3886343	3804877	3777575	3233732	71381	198973	2963378

F2-012	4229760	4098171	4075515	3400396	67522	205765	3127109
F2-014	4402882	4246705	4217462	3474855	84858	208462	3181535
F2-016	4724724	4663953	4637124	4089124	81988	247010	3760126
F2-017	4026781	3902136	3880572	3177891	71559	194659	2911673
F2-018	4463804	4339958	4301512	3638293	85253	211555	3341485
F2-021	4208324	4114691	4093474	3482683	73252	212694	3196737
F2-023	4817911	4724169	4696279	4111942	82048	248662	3781232
F2-024	4051610	3945349	3918127	3433799	75988	209604	3148207
F2-027	3447394	3370581	3352148	2933759	59702	178870	2695187
F2-028	3176883	3092081	3075301	2563545	57098	154385	2352062
F2-029	2072665	2044754	2033550	1881110	37355	114692	1729063
F2-031	4654079	4563601	4530674	3917124	92305	240178	3584641
F2-032	4513564	4427132	4400931	3745045	78885	231875	3434285
F2-033	3905468	3821753	3799933	3278099	68912	201723	3007464
F2-034	4143313	3944454	3912042	3172975	92128	202881	2877966
F2-035	4460515	4348797	4286681	3589512	109985	218489	3261038
F2-036	3638850	3581208	3562153	3127581	65801	192331	2869449
F2-037	3104063	3057920	3040801	2543523	54724	149787	2339012
F2-038	3791592	3702261	3681337	3109942	75636	197033	2837273
F2-039	4079809	3991156	3963856	3316883	78717	206272	3031894
F2-040	4359244	4277014	4252826	3729333	78080	228725	3422528
F2-041	4424904	4342415	4309735	3770599	101416	234599	3434584
F2-042	3197019	3151296	3130606	2708427	59121	169248	2480058
F2-043	3406467	3358337	3336437	2918500	62577	175761	2680162
F2-044	3951385	3899369	3870935	3339555	76469	201814	3061272

F2-045	6284404	6191477	6158362	5354770	114138	330898	4909734
F2-046	4410011	4338718	4304376	3628238	89056	223218	3315964
F2-047	4656184	4572072	4529721	3825451	100972	230230	3494249
F2-048	4235949	4174906	4144009	3604383	82580	220224	3301579
F2-049	3778073	3719151	3696741	3243385	70485	197102	2975798
F2-050	3614402	3552780	3523864	3002737	78206	181074	2743457
F2-051	3921997	3869189	3840054	3382909	80803	206710	3095396
F2-052	3921002	3866829	3839799	3408188	81522	208405	3118261
F2-053	3746924	3679358	3658219	3203554	70043	196857	2936654
F2-054	3099006	2990083	2964600	2345843	61664	148989	2135190
F2-055	3660569	3595850	3574981	2992422	67225	179480	2745717
F2-056	3690586	3623357	3602714	3047193	64749	188326	2794118
F2-057	3717867	3660188	3637928	3173157	70383	193243	2909531
F2-058	4105926	4043927	3999192	3527739	99500	217664	3210575
F2-059	4037483	3941677	3894105	3166723	90409	194493	2881821
F2-060	4255327	4195073	4139938	3667609	108671	222102	3336836
F2-062	2442682	2342097	2324655	1999409	45183	121153	1833073
F2-063	4044055	3421326	3398848	2540145	60151	161852	2318142
F2-065	4562420	4370426	4341320	3672998	77109	223636	3372253
F2-067	4063966	3248016	3226611	2435315	59700	148477	2227138
F2-068	4501078	4291656	4261300	3596536	77414	218010	3301112
F2-069	3763998	3596687	3570021	3055741	68353	189830	2797558
F2-073	3467599	2862909	2844271	2026301	45705	128422	1852174
F2-074	2113129	1879021	1864547	1446810	33495	89031	1324284
F2-075	3279242	2802880	2777906	2089250	55309	127385	1906556

F2-076	3152571	2582099	2553266	1786053	52974	110940	1622139
F2-077	3576745	2978396	2958146	2231023	52907	133738	2044378
F2-081	3822372	3000839	2978207	2142932	54930	133591	1954411
F2-082	3221834	2757149	2739778	1990283	47282	128508	1814493
F2-084	2902882	2823017	2802126	2376148	54180	148589	2173379
F2-085	2834422	2766574	2749175	2372269	48995	146740	2176534
F2-086	4476926	4245141	4211733	3242064	85917	198939	2957208
F2-087	4166207	3784468	3767088	2489581	365301	136768	1987512
F2-090	4307364	4131228	4111373	3038212	101477	190614	2746121
F2-091	5775449	5652845	5612837	4550224	110271	277446	4162507
F2-092	5170680	5043080	5017092	4027777	93948	248742	3685087
F2-094	3502485	3091294	3070077	2132334	72502	135139	1924693
F2-095	3793043	3400338	3381497	2426141	68306	152291	2205544
F2-097	5631697	5473740	5431429	4202993	131041	257041	3814911
F2-100	5552695	5448435	5421581	4434440	182169	274223	3978048
F2-101	5058259	4940847	4914980	3974572	101434	246091	3627047
F2-102	5868673	5750724	5714211	4852383	119656	304152	4428575
F2-104	4245787	4183928	4163073	3528432	90225	216455	3221752
F2-105	3976174	3903723	3883924	3236858	73104	194416	2969338
F2-109	4611294	4508485	4477947	3367207	87277	206980	3072950
F2-112	3935780	3452541	3434027	2396715	62514	147000	2187201
F2-114	4584928	4463304	4439526	3760896	88672	228564	3443660
F2-115	3976727	3645951	3617450	2714776	73610	173101	2468065
F3-002	4732365	4695623	4667220	4227189	93454	260866	3872869
F3-003	4530510	4497438	4446312	4058528	107596	247800	3703132

F3-004	3494958	3465597	3437770	3094675	80426	189267	2824982
F3-005	4008905	3975098	3953886	3603074	80424	222967	3299683
F3-006	3981413	3944865	3921757	3512454	74065	216466	3221923
F3-007	4009902	3934898	3901292	3391506	83756	206844	3100906
F3-008	4203670	4155348	4112736	3714241	96891	223919	3393431
F3-010	3903320	3852583	3827462	3458178	85477	200999	3171702
F3-013	4707809	4637869	4612090	4060792	81563	245650	3733579
F3-015	4004019	3961775	3939006	3465230	82960	210331	3171939
F3-016	4045518	3941408	3897503	3005499	100952	180216	2724331
F3-017	3870350	3803542	3782533	3198222	68393	190079	2939750
F3-018	5289466	5193487	5129398	4450225	134313	270066	4045846
F3-019	4968694	4874184	4840803	4176595	100671	249385	3826539
F3-021	4846642	4760800	4718330	4061734	112608	251649	3697477
F3-022	5485661	5406702	5371432	4761529	103321	287054	4371154
F3-023	4918766	4844293	4804130	4252593	108913	259451	3884229
F3-024	4297543	4237655	4205278	3662540	113518	217671	3331351
F3-025	4125628	4003780	3982539	3291872	75820	206480	3009572
F3-026	4296787	4245407	4212613	3712517	98613	222295	3391609
F3-027	4425671	4229867	4208616	3264656	107091	202465	2955100
F3-028	5600949	5531401	5490581	4889277	131933	293684	4463660
F3-029	4148532	4067676	4031425	3453850	95037	208576	3150237
F3-030	3982090	3895597	3874424	3219679	79762	194441	2945476
F3-031	4587409	4542707	4517905	4094394	101882	248395	3744117
F3-032	4820886	4762210	4721115	4217388	116897	255254	3845237
F3-033	4137313	4070176	4049221	3539499	112810	209973	3216716

F3-034	6932530	6864470	6812048	6126170	149005	375570	5601595
F3-036	4058587	4015749	3993840	3601758	83787	221879	3296092
F3-037	4613312	4529363	4408737	3791647	160293	223043	3408311
F3-038	4493431	4456209	4419331	4055614	104023	246373	3705218
F3-039	4223410	4085029	4052816	3324756	94882	204422	3025452
F3-040	4782023	4727715	4699080	4205465	89578	255451	3860436
F3-041	5104373	5038202	5002847	4397376	109900	258637	4028839
F3-042	5070463	5001922	4975441	4327630	87131	255895	3984604
F3-043	4512205	4433768	4406328	3886462	132652	231826	3521984
F3-044	4828943	4740119	4714472	4079015	85051	242963	3751001
F3-045	4193978	4147239	4123939	3648519	78662	223186	3346671
F3-046	3867138	3818331	3799056	3343681	81888	200284	3061509
F3-047	3248477	2702601	2675439	812411	681679	8795	121937
F3-048	4733353	4676155	4647344	4150695	124297	250201	3776197
F3-049	4178592	4096302	4076007	3280177	76039	199897	3004241
F3-050	1620965	1583421	1573432	1313950	30204	79593	1204153
F3-051	4866046	4787413	4753296	4124830	93591	245847	3785392
F3-052	5382473	5317196	5271560	4715845	123561	285572	4306712
F3-053	5030062	4945004	4918707	4200032	101390	250954	3847688
F3-054	1705134	725423	701312	272447	138394	8510	125543
F3-055	4467490	4422203	4399596	3915017	83162	239868	3591987
F3-056	4290882	4195827	4171100	3360596	80159	196320	3084117
F3-057	4534132	4451430	4428198	3917797	81092	225819	3610886
F3-058	4986279	4911979	4847955	4132730	144699	245002	3743029
F3-060	2966387	2928680	2913737	2327413	107678	137444	2082291

F3-061	4241029	4201901	4178970	3772970	84126	230671	3458173
F3-062	3778495	3740048	3716531	3275658	89340	197066	2989252
F3-071	6236391	6111159	6077506	5089251	251594	295513	4542144
F3-072	4060719	3897180	3868094	3181223	97590	187199	2896434
F3-073	5597613	5511571	5475202	4639427	153004	275064	4211359
F3-074	3589021	3547324	3525685	3019572	92290	180315	2746967
F3-075	3997794	3876374	3853006	3155129	84851	190415	2879863
F3-076	4291383	4248381	4227186	3776363	90117	223036	3463210
F3-077	3289181	3155858	3130438	2456884	73389	146459	2237036
F3-078	4805665	4675807	4638121	3825478	108006	232317	3485155
F3-080	4525942	4361559	4337796	3445493	92337	208492	3144664
F3-081	6354364	6254813	6206422	5282329	161739	312721	4807869
F3-082	4019517	3857867	3830039	3104998	107824	181485	2815689
F3-083	3666566	3395901	3373205	2619468	95789	156053	2367626
F3-084	5485221	5394189	5365678	4562542	133228	275086	4154228
F3-085	3567339	3524531	3495559	3037784	87581	178985	2771218
F3-086	3372036	3224191	3206008	2452585	59842	148306	2244437
F3-087	3750933	3592958	3574644	2824996	104467	166818	2553711
F3-088	4015205	3740143	3719044	2854018	74983	172689	2606346

MOLECULAR ECOLOGY
RESOURCES

Table S5: Statistics of the genetic maps constructed in *Solea senegalensis* using three full-sib families. Maps were constructed via male and via female in each family and the consensus per sex and for the whole species obtained. The correspondence between linkage groups in the consensus map and that reported by Guerrero-Cózar et al. (2021) is also provided. LG codes were arranged from the longest to the shortest within each map, but their correspondence was established according to the codes of the consensus map, which in turn followed the chromosome number of the karyotype after mapping integration. LG: linkage group; families: F1, F2 and F3; male: M; female: F.

**Due to space issues, Table S5 is enclosed as an additional attached file (.xlsx).*

Table S6: Marker positions for all genetic maps constructed in *Solea senegalensis* and their integration by sex and species.

**Due to space issues, Table S6 is enclosed as an additional attached file (.xlsx).*

MOLECULAR ECOLOGY
RESOURCES

Table S7. List of anchored contigs of the *Solea senegalensis* genome on the genetic map indicating the orientation and size.

LG / chrom	pseudo-chromosomes*	Scaffold	Orientation	size (bp)	size /chrom
1	1	SolSen1_s38c1	Reverse	781045	46363013
		SolSen1_s13c1	Reverse	23439045	
		SolSen1_s15c1	Forward	22142923	
2	3	SolSen1_s23c1	Reverse	13730948	36524923
		SolSen1_s30c1	Reverse	3794735	
		SolSen1_s20c1	Reverse	18999240	
3	16	SolSen1_s10c1	Reverse	24069211	24356113
		SolSen1_s56c1	Forward	286902	
4	2	SolSen1_s46c1	Forward	439503	39681215
		SolSen1_s07c1	Reverse	24577862	
		SolSen1_s22c1	Reverse	14663850	
5	21	SolSen1_s14c1	Reverse	22452756	22452756
6	5	SolSen1_s42c1	Forward	522248	30211065
		SolSen1_s01c1b	Reverse	21630749	
		SolSen1_s28c1	Reverse	8058068	
7	7	SolSen1_s09c1	Reverse	24206605	33076213
		SolSen1_s43c1	Reverse	507339	
		SolSen1_s27c1	Reverse	8362269	
8	10	SolSen1_s04c1	Reverse	28132043	28132043
9	12	SolSen1_s45c1	Reverse	446947	27888067
		SolSen1_s29c1	Forward	5975391	
		SolSen1_s01c1a	Reverse	12020340	
		SolSen1_s25c1	Reverse	8875931	
		SolSen1_s41c1	Forward	569458	
10	20	SolSen1_s37c1	Forward	856747	22931278
		SolSen1_s48c1	Forward	395635	
		SolSen1_s16c1	Reverse	20937294	
		SolSen1_s47c1	Forward	414049	
		SolSen1_s50c1	Forward	327553	
11	17	SolSen1_s12c1	Forward	23564453	23564453
12	18	SolSen1_s33c1	Reverse	2416913	

		SolSen1_s19c1	Reverse	19354633	21771546
13	11	SolSen1_s18c1	Forward	19885849	
		SolSen1_s26c1	Reverse	8756652	28642501
14	6	SolSen1_s52c1	Forward	317263	
		SolSen1_s64c1	Forward	224809	
		SolSen1_s24c1	Reverse	13270792	
		SolSen1_s21c1	Reverse	15211903	29024767
15	19	SolSen1_s32c1	Reverse	3259233	
		SolSen1_s17c1	Reverse	20639214	
		SolSen1_s36c1	Forward	889564	24788011
16	9	SolSen1_s05c1	Reverse	27800896	27800896
17	13	SolSen1_s63c1	Forward	229283	
		SolSen1_s11c1	Forward	23966579	24195862
18	15	SolSen1_s08c1	Reverse	24552006	24552006
19	4	SolSen1_s35c1	Reverse	1032886	
		SolSen1_s02c1	Reverse	30103958	31136844
20	8	SolSen1_s34c1	Forward	1773698	
		SolSen1_s03c1	Reverse	28862912	30636610
21	14	SolSen1_s06c1	Forward	25475828	
		SolSen1_s31c1	Forward	3674203	29150031
Total				606880213	

* Pseudochromosomes from Guerrero-Cózar et al. (2021)

MOLECULAR ECOLOGY
RESOURCES

Table S8. Comparative statistics of the *Solea senegalensis* genome with other pleuronectiform chromosome-level genomes.

	Solea senegalensis		***Platychthys stellatus***	***Scophthalmus maximus***	***Cynoglossus semilaevis***	***Hippoglossus hippoglossus***	***Hippoglossus stenolepis***	***Reinhardtius hippoglossoides***	***Verasper variegatus***	***Paralichthys olivaceous***
	this study	***Guerrero-Cózar et al.* (2021)**	Lü et al (2021)	Martínez et al. (2021)	Chen et al (2014)	Einfeldt et al. (2021)	Jasonowicz et al. (2022)	Ferchaud et al., 2022	Zhao et al., 2021	Shao et al., 2017
Chromosome number	21	21	24	22	21	24	24	24	23	24
Bioproject		PRJNA643826	PRJNA592732	PRJNA631898	PRJNA251742	PRJNA562001	PRJNA622249	PRJNA499109	PRJNA634516	PRJNA73673
GC_content (%)	40.90	40.92	42.66	43.38	41.27	42.27	42.24	42.65	42.11	41.62
Contig / scaffold N50 (Mb)	28.6	26	25.1	25.94	0.5	26.31	27.29	25.01	24.76	3.81
Largest contig / scaffold (Mb)	36.20	42.92	31.75	32.11	NA	31.67	32.85	31.98	31.93	NA
Total length (Mb)	613.0	603.53	609.99	556.7	470.19	596.79	602.15	598.52	545.34	545.77
% assembled in chromosomes	99%	90%	92%	96%	94%	94%	99.8%	96%	99%	98%
Contig / scaffold number	81	1,937	31,621	127	31,181	57	52	1590	57	7,202

MOLECULAR ECOLOGY RESOURCES

Table S9: One hundred and forty-one BAC clones used for integrating cytogenetic, genetic and physical maps in *Solea senegalensis*. Notice that some clones highlighted in bold matched to several genomic regions, either in different or the same chromosome, among them, the 5S rDNA gene clusters.

Chrom.	BAC	start	end	Size(bp)	References
1	13G1	7572042	7580904	8862	García-Angulo et al. 2018
1	10F5	7744068	7856487	112419	Ramírez et al., 2022
1	67N4	12105594	12350291	244697	Ramírez et al., 2022
1	48P7	16598420	16840821	242401	García-Angulo et al. 2018; Rodríguez et al. 2019
1	67P21	18262950	18472071	209121	Ramírez et al., 2022
1	74O9	19014762	19198467	183705	Ramírez et al., 2022
1	12D22	19192044	19237400	45356	Merlo et al. 2017
1	52C17	30927821	31108578	180757	García-Angulo et al. 2018; Rodríguez et al. 2019
1	10K23	35270646	35377832	107186	Portela-Bens et al. 2017
1	73B7	35278957	35318620	39663	García-Angulo et al. 2018; Rodríguez et al. 2019
1	36D3	37696959	37745328	48369	García-Angulo et al. 2018
1	5K5	44589098	44763667	174569	Merlo et al. 2017
1	10L10	44615210	44668115	52905	García-Angulo et al. 2018
2	19J21	5513333	5563516	50183	Portela-Bens et al. 2017
2	68P5	12794656	12801151	6495	Ramírez et al., 2022
2	38N10	15120480	15330995	210515	García-Angulo et al. 2019
2	21I14	18113402	18183817	70415	Ramírez et al., 2022
2	36K1	18293923	18317370	23447	García et al. 2019
2	21O23	22006794	22057985	51191	Portela-Bens et al. 2017

2	36I3	24726080	24752892	26812	García et al. 2019
2	65E23	24977297	25001291	23994	Ramírez et al., 2022
2	46C5	31008363	31187699	179336	García et al. 2019
2	60P19	31330275	31500970	170695	García-Angulo et al. 2020
2	4D15	32094905	32160473	65568	García-Angulo et al. 2019
2	42D4	32859458	33012486	153028	Ramírez et al., 2022
2	52G10	33722667	33895699	173032	García-Angulo et al. 2019
3	31O1	1492949	1508353	15404	Ramírez et al., 2022
3	54H18	1683791	1884991	201200	Ramírez et al., 2022
3	9C12	6222465	6426406	203941	Ramírez et al., 2022
3	**59B23**	7901674	7994798	93124	Merlo et al. 2021
3	**73A11**	15686835	15865315	178480	Ramírez et al., 2022
3	4B13	17427128	17540031	112903	Ramírez et al., 2022
3	9J4	19777023	19917354	140331	García et al. 2019
4	36H2	639777	700858	61081	García et al. 2019
4	36H3	639785	691944	52159	García et al. 2019
4	36J2	2001465	2053869	52404	García et al. 2019
4	8A23	6221120	6315616	94496	García et al. 2019
4	67K3	9638913	9728429	89516	Ramírez et al., 2022
4	12D24	15417518	15590547	173029	Arias-Pérez et al. 2018
4	**30J4**	15964546	16159640	195094	Cegarra et al. 2013; Merlo et al. 2017¸ García-Angulo et al. 2018
4	46B2	16297804	16497587	199783	García et al. 2019
4	3C15	16826575	16895274	68699	García et al. 2019
4	30P17	21467452	21552765	85313	García et al. 2019
4	12N15	32339370	32494473	155103	Portela-Bens et al. 2017
4	39G22	3603742	3624251	205087	Ramírez et al., 2022

		4	1		
5	74M4	7197228	7271114	73886	Ramírez et al., 2022
5	**72B11**	7198891	7268749	69858	Ramírez et al., 2022
5	46K16	13507583	13702555	194972	Merlo et al. 2021
5	3I18	16307359	16495687	188328	Ramírez et al., 2022
6	**64A8**	1625716	1666503	40787	Merlo et al. 2021
6	47B18	5076008	5082364	6356	Ramírez et al., 2022
6	20D18	5324057	5504651	180594	Portela-Bens et al. 2017
6	11O20	5438136	5525880	87744	Portela-Bens et al. 2017
6	**53B20**	6919675	7149217	229542	Rodríguez et al. 2019
6	31A1	7712533	7789363	76830	García et al. 2019
6	**5SrDNA**	9659398	9672188	12790	Cross et al. 2008
6	**64A8**	15270928	15434386	163458	Merlo et al. 2021
6	67P7	15441751	15642191	200440	Ramírez et al., 2022
6	48K7	16397982	16574387	176405	García-Angulo et al. 2018; Rodríguez et al. 2019
6	16E17	16505513	16549247	43734	Portela-Bens et al. 2017
6	**53B20**	16796598	16976225	179627	Rodríguez et al. 2019
6	**5SrDNA**	17148051	17151768	3717	Cross et al. 2008
6	**45M23**	29822138	29834935	12797	Merlo et al. 2021
7	39D10	2480478	2665831	185353	Ramírez et al., 2022
7	**44K21**	3983841	4039110	55269	Ramírez et al., 2022
7	19H19	4557359	4620134	62775	Portela-Bens et al. 2017
7	47G8	6288866	6330779	41913	Ramírez et al., 2022
7	76F9	11592874	11746665	153791	Ramírez et al., 2022
7	13O12	11721375	11934095	212720	Ramírez et al., 2022
7	7H22	20728761	20788681	59920	Portela-Bens et al. 2017
8	31A2	4121067	4191789	70722	García et al. 2019
8	8O7	10371887	10534927	163040	Potela-Bens et al. 2017

8	57C10	11166674	11359763	193089	Ramírez et al., 2022
8	**57G6**	12350077	12393163	43086	Merlo et al. 2021
9	51E11	3089803	3103902	14099	Ramírez et al., 2022
9	9B2	6516733	6659604	142871	Ramírez et al., 2022
9	4N9	6980991	7147440	166449	Ramírez et al., 2022
9	57G16	10929122	10951606	22484	Merlo et al. 2021
9	**72B11**	10932926	10951606	18680	Ramírez et al., 2022
9	39F2	13409737	13499914	90177	García-Angulo et al. 2020
9	32B8	19773666	19969345	195679	Portela-Bens et al. 2017
10	15I19	5671172	5879470	208298	García et al. 2019
10	**68G4**	6293356	6440901	147545	Merlo et al. 2021
10	**57N7**	6612611	6660809	48198	Ramírez et al., 2022
10	**68G4**	7646186	7701312	55126	Merlo et al. 2021
10	9E8	8417647	8456718	39071	García-Cegarra et al. 2013
10	13L18	15925939	16130724	204785	Ramírez et al., 2022
10	56H24	16109115	16239502	130387	García-Angulo et al. 2018
11	38F24	8094847	8147905	53058	García-Angulo et al. 2020
11	3N10	8177480	8232219	54739	Merlo et al. 2021
11	31F1	9037817	9083045	45228	García et al. 2019
11	4E10	12859860	12924476	64616	García et al. 2019
11	**45L11**	12873354	13066645	193291	Merlo et al. 2021
11	**72B11**	16071434	16149877	78443	Ramírez et al., 2022
11	**5SrDNA**	22649871	22697902	48031	Cross et al. 2008
12	13F2	1252582	1258497	5915	García et al. 2019
12	38B21	10317182	10493398	176216	Ramírez et al., 2022
12	13E1	12782153	12805894	23741	García-Cegarra et al. 2013
12	**45M23**	13193012	13227985	34973	Merlo et al. 2021

12	39L15	13269511	13457052	187541	Ramírez et al., 2022
12	**57N7**	14614233	14714712	100479	Ramírez et al., 2022
12	35D17	15652773	15764583	111810	García et al. 2019
12	**30J4**	19124482	19282990	158508	Cegarra et al. 2013; Merlo et al. 2017¸ García-Angulo et al. 2019
13	19L16	15543458	15555778	12320	Merlo et al. 2021
13	71J13	16353484	16581621	228137	Merlo et al. 2021
13	51K20	16903092	17096081	192989	Ramírez et al., 2022
13	65J17	19342693	19489797	147104	Ramírez et al., 2022
13	66D13	21411668	21632941	221273	Ramírez et al., 2022
13	4M14	23145203	23249840	104637	García-Angulo et al. 2020
14	**57G6**	9950581	10048237	97656	Merlo et al. 2021
14	**73A11**	10248677	10337286	88609	Ramírez et al., 2022
14	4N21	12880781	12890280	9499	Ponce et al. 2011
14	58D1	13131338	13309164	177826	Ramírez et al., 2022
14	29D4	23017672	23212382	194710	García-Angulo et al. 2019
15	36E3	523083	532570	9487	García et al. 2019
15	22C2	3403056	3427067	24011	García-Cegarra et al. 2013
15	60P24	12187693	12241663	53970	Merlo et al. 2021
15	4F12	17969998	18029401	59403	García et al. 2019
15	**44K21**	21276075	21432848	156773	Ramírez et al., 2022
16	71N11	485078	648699	163621	García-Angulo et al. 2019
16	53D20	3651069	3763845	112776	García-Angulo et al. 2019
16	54E19	9764895	9870163	105268	Ramírez et al., 2022
16	25P16	11149350	11205906	56556	García-Angulo et al. 2020

16	52E18	22853786	23047053	193267	García et al. 2019
16	**57G6**	23915869	24003315	87446	Merlo et al. 2021
16	76A22	26503639	26528965	25326	Ramírez et al., 2022
16	**57N7**	26503640	26524778	21138	Ramírez et al., 2022
17	42F9	3430436	3627436	197000	Ramírez et al., 2022
17	**53B20**	5770620	5877014	106394	Rodríguez et al. 2019
17	65I16	10312738	10492084	179346	Merlo et al. 2021
17	19K18	10334158	10465773	131615	Merlo et al. 2021
17	65O21	12557319	12669357	112038	Ramírez et al., 2022
17	73J17	17132878	17167492	34614	Merlo et al. 2021
17	63A7	19159174	19346742	187568	García-Cegarra et al. 2013
17	31N1	23697949	23769497	71548	García et al. 2019
18	**45L11**	2053773	2098772	44999	Merlo et al. 2021
18	36M2	16710945	16782020	71075	García et al. 2019
18	3F15	17971067	18091131	120064	Merlo et al. 2021
18	15B1	19115653	19305121	189468	Ramírez et al., 2022
18	2K18	24213324	24266017	52693	Portela-Bens et al. 2017
19	31C1	96421	256360	159939	García-Angulo et al. 2019
19	38H3	2348678	2363954	15276	Ramírez et al., 2022
19	58D11	3537958	3718661	180703	Ramírez et al., 2022
19	50K3	11941263	12174712	233449	García-Angulo et al. 2020
19	13F4	17584531	17669759	85228	García et al. 2019
19	42P4	22828847	23030118	201271	García-Angulo et al. 2019
19	54G7	23013113	23235840	222727	Ramírez et al., 2022

19	62G15	23242376	23289156	46780	Ramírez et al., 2022
19	**45L11**	23651933	23679580	27647	Merlo et al. 2021
19	12K16	26368643	26377777	9134	García-Cegarra et al. 2013
20	45M19	3025095	3104382	79287	Ramírez et al., 2022
20	53K8	13442048	13625281	183233	Ramírez et al., 2022
20	54F6	17844694	18069920	225226	Merlo et al. 2021
20	1C2	25751479	25790726	39247	García-Angulo et al. 2018
21	63A3	4375986	4539254	163268	García-Cegarra et al. 2013; García et al. 2019
21	**59B23**	17941401	18146632	205231	Merlo et al. 2021
21	55B12	20402371	20446655	44284	García-Angulo et al. 2020
21	3A12	20787667	20968427	180760	Ramírez et al., 2022
21	72O12	21763890	21985271	221381	Ramírez et al., 2022
21	30H22	23487358	23593948	106590	Portela-Bens et al. 2017

MOLECULAR ECOLOGY
RESOURCES

Table S10: Wright F-statistics for male and female populations per SNP and using 50 SNP-sliding windows averaged over F_{IS} and F_{ST} across chromosome 12 of *Solea senegalensis* (between 9,371719 and 11,071,605) bp, where the *fshr* gene is located. Highlighted in green the region of maximum F_{ST} and minimum F_{IS} values.

**Due to space issues, Table S10 is enclosed as an additional attached file (.xlsx).*

Table S11: SNPs localized in the follicule stimulating hormone receptor (*fshr*) gene in six males and six females resequenced at 20x coverage using the *Solea senegalensis* assembled genome; diagnostic SNPs are homozygous in females and heterozygous in males consistent with a XX / XY sex determining system; in the last row it is indicated if SNPs determine aminoacid substitutions (non-synonymous) or not (synonymous); the boundaries of exons are highlighted in bold type.

**Due to space issues, Table S11 is enclosed as an additional attached file (.xlsx).*

Table S12. Sets of primers designed with Primer 3 to develop a molecular tool for sexing in *Solea senegalensis* using diagnostic markers between males (heterozygous) and females (homozygous) located at exon 14 of the *fshr* gene.

**Due to space issues, Table S12 is enclosed as an additional attached file (.xlsx).*

MOLECULAR ECOLOGY
RESOURCES

Table S13: Genotypes and allelic counts of diagnostic SNPs located at the *fshr* gene (exons, 5' and 3' UTR, introns; detailed information in Table S14) from gonad RNAseq data of samples of five males (M) and five females (F) collected across gonad development of *Solea senegalensis*, from the initial undifferentiated or low differentiated stages (84D, 98D and 126D post fertilization) until juveniles and adults; SNP ID makes reference to the contig and position where they are located in the contig of the genome assembly; REF and ALT alleles refers to the allele in the genome (0) and the alternative allele (1) detected after resequencing six females and six males; Genotypes: homozygous in females (0/0 or 1/1) and heterozygous in males (0/1); ./. missing genotypes because allelic counts did not reach the minimum threshold (8 reads); allelic counts: the first number refers to the "0" allele and the second, separated by semicolon, to "1" allele; Colors: pink (missing genotypes), green (valid genotypes for counting), purple (females); blue (males); red (individuals not considered because they did not reach a minimum genotyping data).

**Due to space issues, Table S13 is enclosed as an additional attached file (.xlsx).*

MOLECULAR ECOLOGY
RESOURCES

Table S14. Quality assessment of modeled Y-linked and X-linked allelic variants of the *fshr* SD gene of *Solea senegalensis.*

		Procheck				C-score*	Confidence score**
		core (%)	***allow (%)***	***gener (%)***	***disall (%)***		
Y-linked	TASSER	68.1	26.3	3.2	2.5	-1.62	-
	ROBETTA	86.5	11.7	1.0	0.8	-	0.7405
X-linked	TASSER	65.7	26.8	5.0	2.5	-2.08	-
	ROBETTA	86.8	11.3	0.8	1.0	-	0.7445

* Confidence score of the I-TASSER algorithm for the estimation of predicted model's quality. Typically ranges between -5 and 2. Higher values means a model with a high confidence and vice-versa.

** Confidence scores as calculated by ROBETTA for estimating the quality of the predicted models. It ranges from 0 to 1, indicating bad and good model's quality, respectively.

MOLECULAR ECOLOGY
RESOURCES

2. SUPPLEMENTARY FIGURES

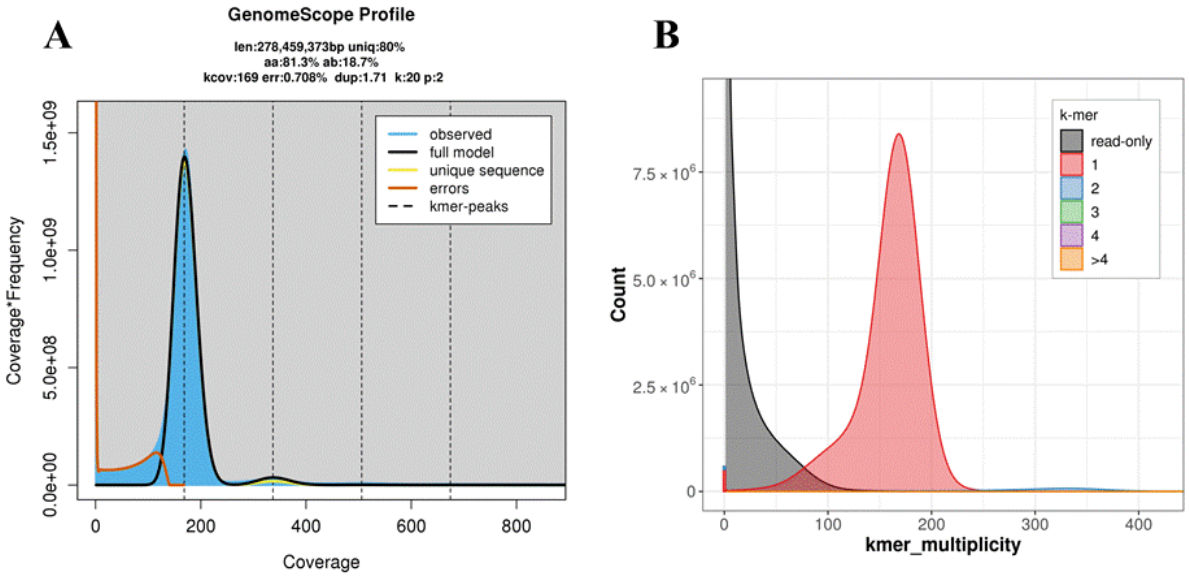


Fig. S1: K-mer distribution on: A) initial Illumina reads, B) the final assembly of *Solea senegalensis*.

MOLECULAR ECOLOGY
RESOURCES

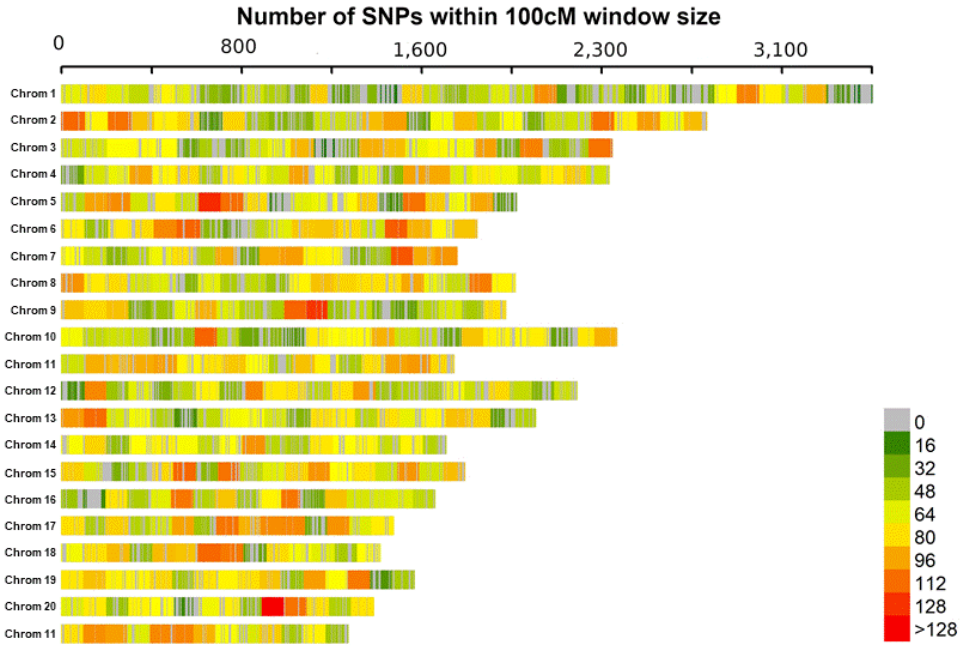


Fig. S2: Mapping marker density across linkage groups in the *Senegalese sole* consensus map.

MOLECULAR ECOLOGY
RESOURCES

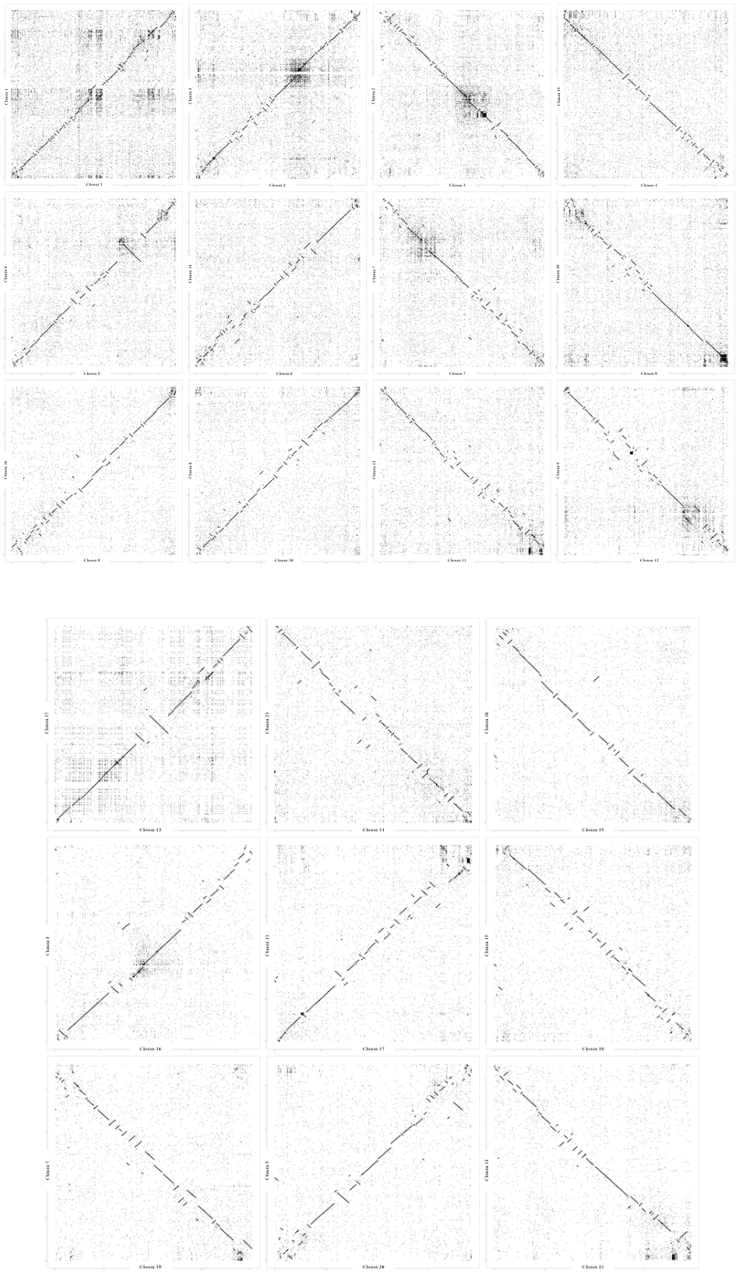


Fig. S3: LASTZ plots between scaffolds / chromosomes (this study; ordinates) and pseudo-chromosomes (Guerrero-Cózar et al., 2021; abscissa) of *Solea senegalensis* genome assemblies; A) psedochromosomes 1-12; B) pseudochromosomes 13-21.

MOLECULAR ECOLOGY
RESOURCES

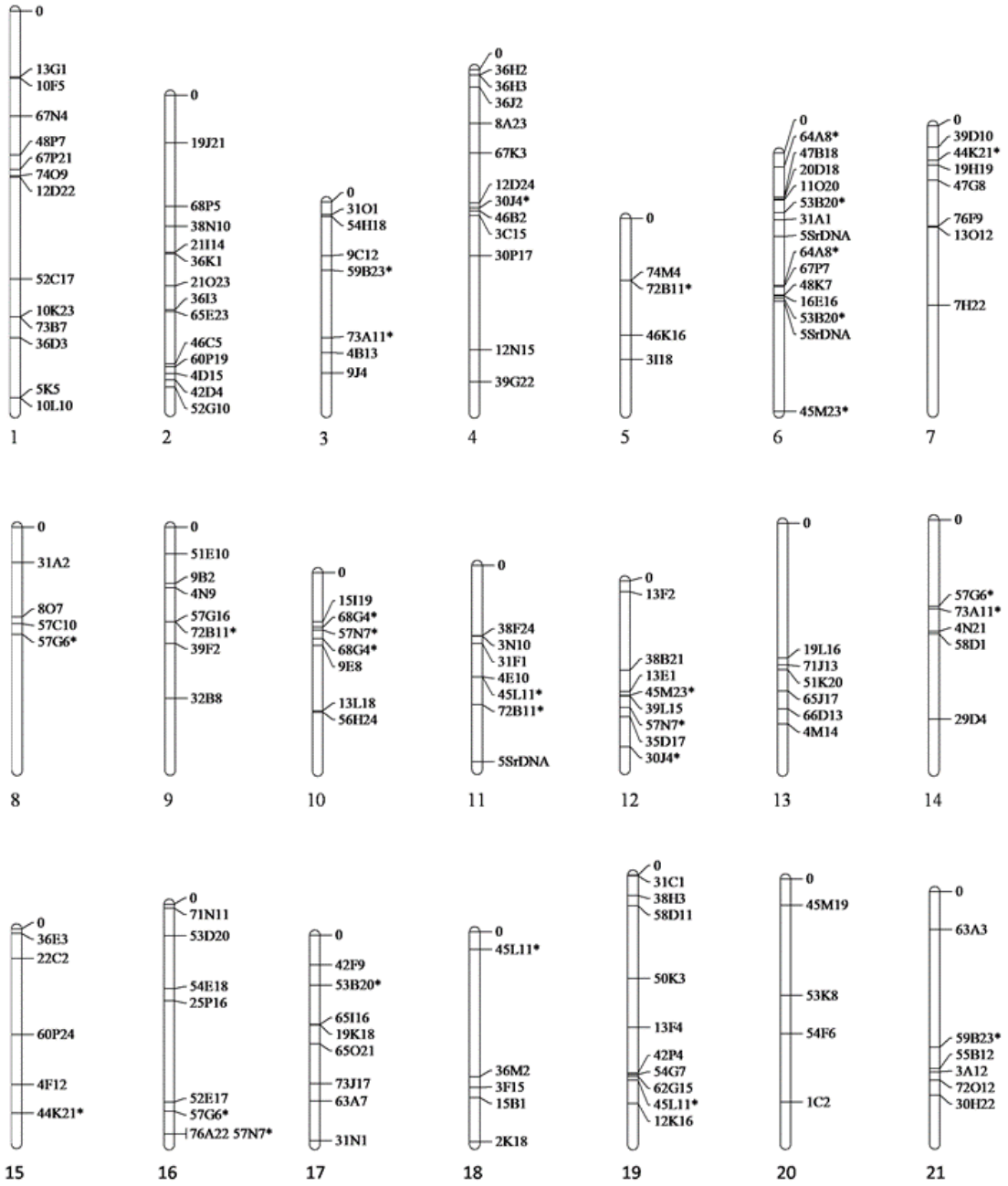


Fig. S4: Idiogram of the *Solea senegalensis* chromosomes where position of BACs used for mapping integration is shown.

MOLECULAR ECOLOGY
RESOURCES

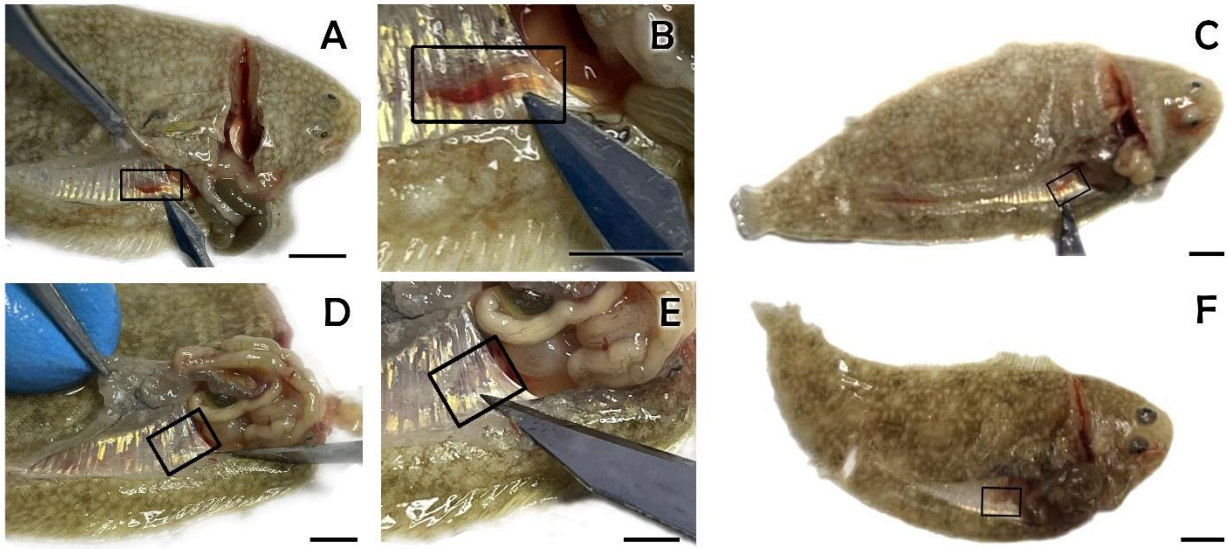


Fig. S5:. Macroscopic anatomy and topography of *Solea senegalensis* gonads of individuals of 126, 98 and 84 dpf. (A, B) 126 dpf female sole; (C) 98 dpf individual - unidentified sex; (D, E) 126 dpf presumably sole male identified by 'lacking a female gonad'. (F) 84 dpf individual with unidentified sex. Insets: site of gonad. Scale bars: 600 µm (A); 400 µm (B-F).

**Due to space issues, Fig. S6 is enclosed as an additional attached file (.jpg).*

Fig. S6: Histological sections of adult and juvenile *Solea senegalensis* gonads. (A-C) Adult female. (D-F) Juvenile female. (G-I) Adult male. (J-L) Juvenile male. (A-F) (*) Atresia stages; (^) Post-ovulatory follicles; Arrow: Nuclei; Arrowhead: Nucleolus; (1) Oogonia; (2) Early oocyte; (3) Late oocyte. (G-L) White arrows: Show the radial disposition of seminiferous lobules (*) from the central medulla (m) to the cortex (c) and tunica albuginea (ta); (1) and black arrowhead: Spermatids; 2 and black arrow: Spermatozoa; Black square: Interstitial tissue. Stain: Hematoxilin-Eosin (HE). Scale bars: 250 µm (A, D, G); 100 µm (B, C, E, H, J, K); 50 µm (F, I, L).

**Due to space issues, Fig. S7 is enclosed as an additional attached file (.jpg).*

Fig. S7: Histological sections of female gonad of *Solea senegalensis* individuals of 126, 98 and 84 dpf. (A) In 126 dpf female sole, previtellogenic oocytes can be visualized; (B, C) Gonad of 98 and 84 dpf females, respectively, both undifferentiated, although at 98 dpf there are more potential-oocyte-cells. (D-F) Higher magnification of A-C, respectively. Gonads of 98 and 84 dpf individuals correspond to females identified with the SS-sex. Stain: HE. Scale bars: 250 µm (A); 100 µm (B); 50 µm (C, D, E, F).

**Due to space issues, Fig. S8 is found as an additional attached file (.jpg).*

MOLECULAR ECOLOGY
RESOURCES

Fig. S8: Histological sections of male gonad of individuals of 126, 98 and 84 dpf. (A) 126 dpf; (B) 98 dpf; (C) 84 dpf; (D, E) Higher magnification of A, B, respectively. The three stages are undifferentiated, with the oldest one (126 dpf) showing more potentially-spermatogonia cells. All samples were genotyped with the SS-sex marker. * Gonad; K: kidney; c: Cartilage. Stain: HE. Scale bars: 100 µm (A, B, C); 50 µm (D, E).

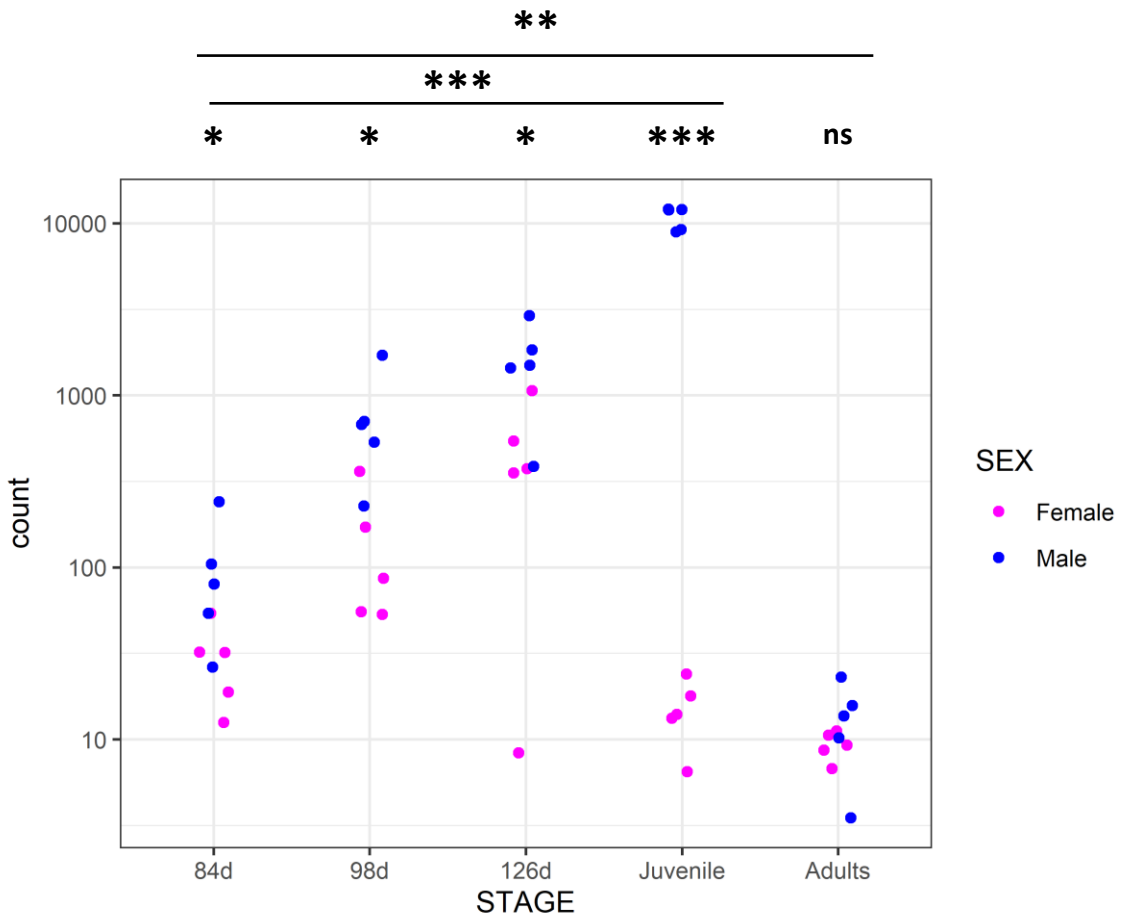
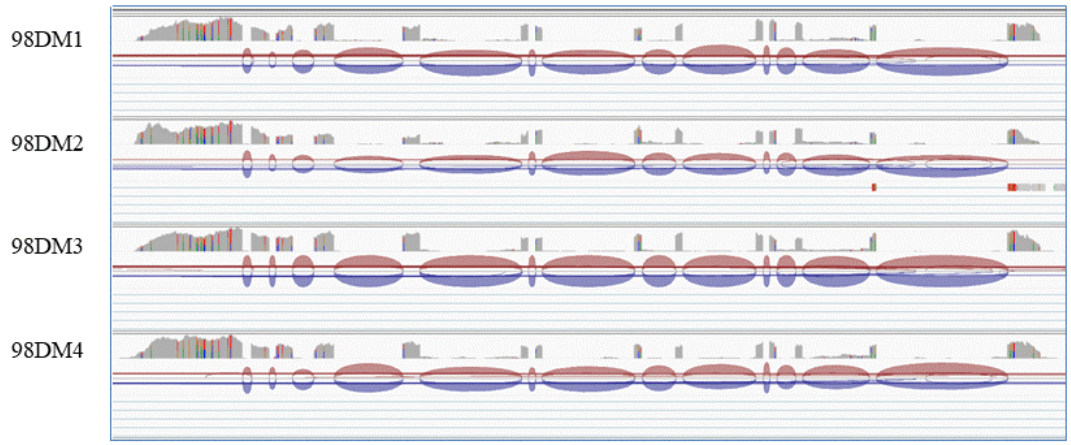


Fig. S9: RNA-SeqRNA-Seq data of the *fshr* gene in *Solea senegalensis* males and females across different gonad developmental stages (abscissas) using a count log scale (ordinates).

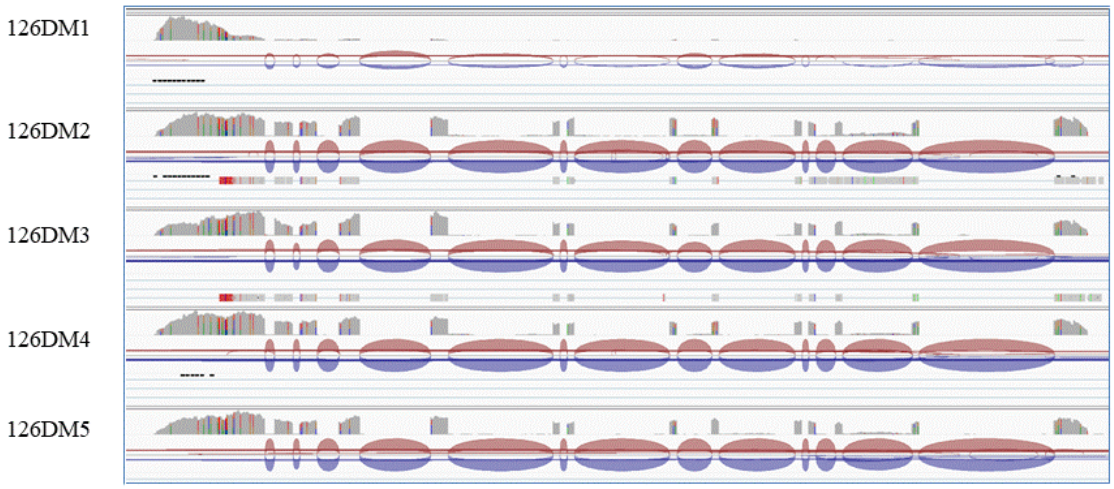
MOLECULAR ECOLOGY
RESOURCES

Mann-Whitney tests for differences between X-linked and Y-linked alleles: * P < 0.05, ** P < 0.01, *** P < 0.001

MALES 126 dpf



MALES 126 dpf



MOLECULAR ECOLOGY
RESOURCES

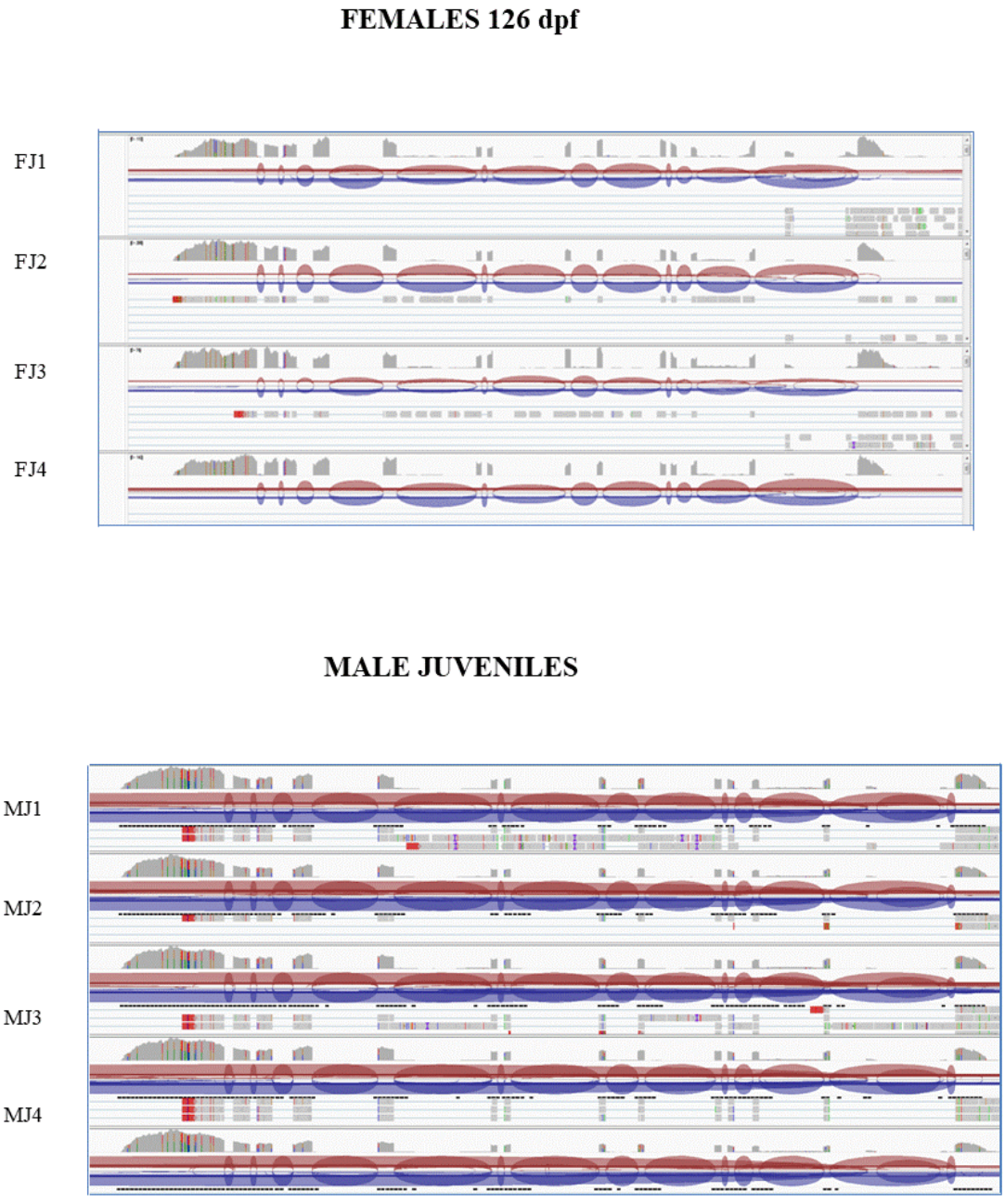


Fig. S10: Read distribution across the 14 exons of the *fshr* gene in males and in females of *Solea senegalensis* at some stages with a number of counts high enough to assess differences.

MOLECULAR ECOLOGY
RESOURCES

Fig. S11: Phylogeny of the *fshr* gene in Pleuronectiformes including the Y- and X-linked allelic variants of *Solea senegalensis* using *D. rerio* as outgroup. Confidence bootstrapping values of each grouping are shown in brackets at the nodes and genetic distances to the nodes above each branch or in parentheses in the short terminal branches.

MOLECULAR ECOLOGY
RESOURCES

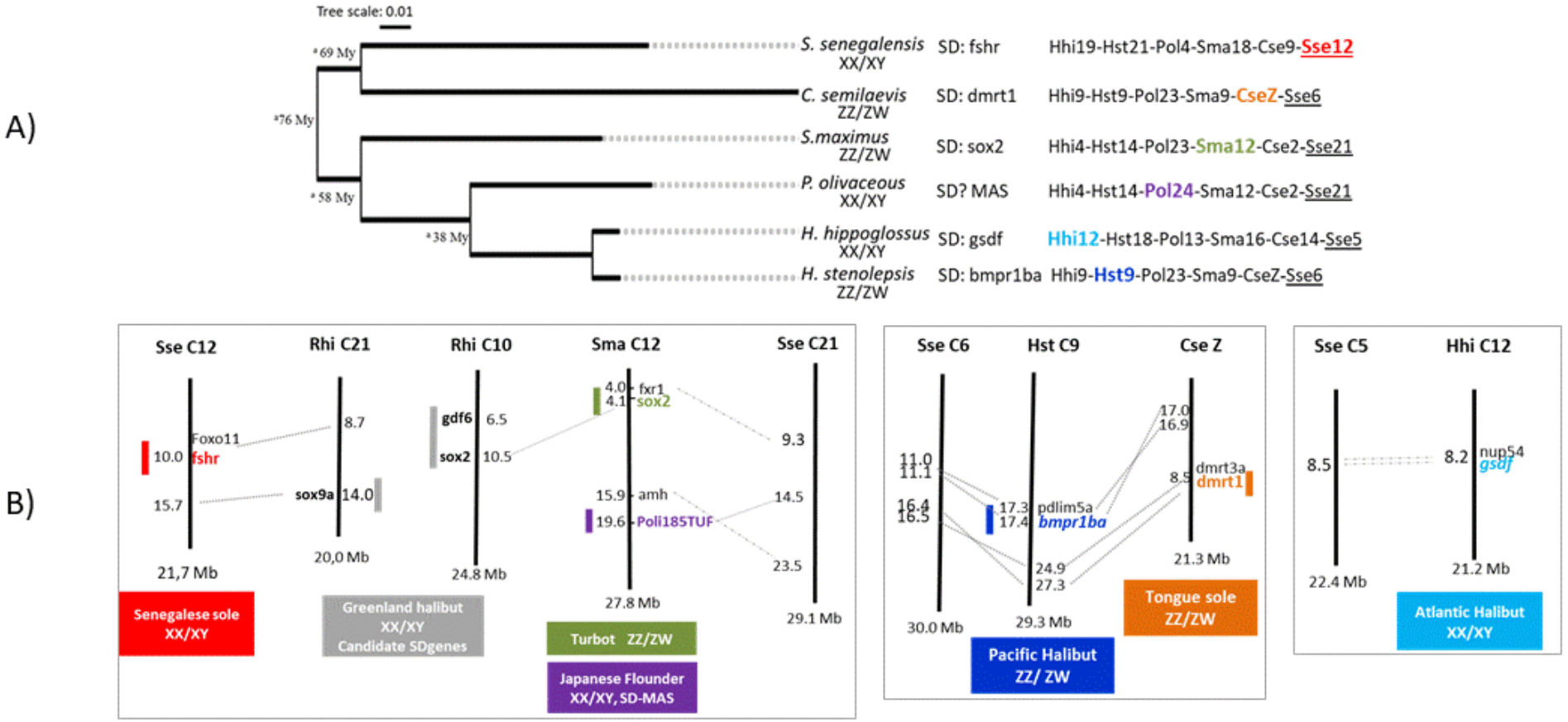


Fig. S12: Diversification of the sex determinant (SD) systems across Pleuronectiformes.A) Concatenated synteny of SD chromosomes and genes (or markers / MAS) in the six flatfish species studied ([a] Phylogenomic divergence (million years, My) among flatfish

families; Lü et al. 2011); B) Comparative mapping between flatfish SD-gene bearing chromosomes. Species/assembly codes and references: Atlantic halibut, *Hippoglossus hippoglossus* (Hhi; fHipHip1.pri; GCA_009819705.1; Edvardsen et al. 2022; Einfeldt et al. 2021); Pacific halibut, *H. stenolepsis* (Hst; UL_REHI_2.0; GCA_006182925.3; Jasonowicz et al., 2022); Japanese flounder, *Paralychthys olivaceous* (Pol; *P*arOli_1.1; GCA_001904815.2). SD-MAS (SD-marker assisted selection), https://patentscope.wipo.int/search/en/detail.jsf?docId=WO2008117715a; Turbot, *Scophthalmus maximus* (Sma; ASM1334776v1; GCA_013347765.1; Martínez et al., 2020); Tongue sole, *Cynoglossus semilaevis* (Cse; Cse_v1.0; GCA_000523025.1; Chen et al. 2014)*;* Senegalese sole, *Solea senegalensis* (Sse; fSolSen1.1_lg; GCA_919967415.2; THIS STUDY); **Greenland halibut (*Reinhardtius hippoglossoides; Rhi*;** UL_REHI_2.0; GCA_006182925.3; Ferchaud et al. 2022).

References:

Arias-Pérez, A., Ramírez, D., Rodríguez, M. E., Portela-Bens, S., García, E., Merlo, M. A., García-Angulo, A., Cross, I., Liehr, T., & Rebordinos, L. (2018). In silico detection and FISH analysis to determine location of miRNAs in *Solea senegalensis* chromosomes using BACs. *OBM Genetics*, *2*, 44. https://doi.org/10.21926/obm.gemmsenet.1804044

Cross, I., Merlo, A., Manchado, M., Infante, C., Cañavate, J., & Rebordinos, L. (2008). Cytogenetic characterization of the sole *Solea senegalensis* (Teleostei: Pleuronectiformes: Soleidae): Ag-NOR, (GATA)n, (TTAGGG)n and ribosomal genes by one-color and two-color FISH. *Genetica*, *128*, 253–259. https://doi.org/10.1007/s10709-005-5928-9

García, E., Cross, I., Portela-Bens, S., Rodríguez, M., García-Angulo, A., Molina, B., Cuadrado, A., Liehr, T., & Rebordinos, L. (2019). Integrative genetic map of repetitive DNA in the sole *Solea senegalensis* genome shows a rex transposon located in a proto-sex chromosome. *Scientific Reports*, *9*, 17146. https://doi.org/10.1038/s41598-019-53673-6

García-Angulo, A., Merlo, M., Iziga, R., Rodríguez, M., Portela-Bens, S., Al-Rikabi, A., Liehr, T., & Rebordinos, L. (2020). Gene clusters related to metamorphosis in *Solea senegalensis* are highly conserved. 2020. *Comparative Biochemistry and Physiology Part D: Genomics and Proteomics*, *35*, 100706. https://doi.org/10.1016/j.cbd.2020.100706

García-Angulo, A., Merlo, M., Portela-Bens, S., Rodríguez, M., García, E., Al-Rikabi, A., Liehr, T., & Rebordinos, L. (2018). Evidence for a Robertsonian fusion in *Solea senegalensis* (Kaup, 1858) revealed by zoo-FISH and comparative genome analysis. *BMC Genomics*, *19*, 818. https://doi.org/10.1186/s12864-018-5216-6

García-Angulo, A., Merlo, M., Rodríguez, M., Portela-Bens, S., Liehr, T., & Rebordinos, L. (2019). Genome and phylogenetic analysis of genes involved in the immune system of *Solea senegalensis* – Potential applications in aquaculture. *Frontiers in Genetics*, *11*, 529. https://doi.org/10.3389/fgene.2019.00529

Merlo, M., Iziga, R., Portela-Bens, S., Cross, I., Kosyakova, N., Liehr, T., Manchado, M., & Rebordinos, L. (2017). Analysis of the histone cluster in *Senegalese sole* (*Solea senegalensis*): Evidence for a divergent evolution of two canonical histone clusters. *Genome*, *60*, 441–453. https://doi.org/10.1139/gen-2016-0143

Merlo, M., Portela-Bens, S., Rodríguez, M., García-Angulo, A., Cross, I., Arias-Pérez, A., García, E., & Rebordinos, L. (2021). A comprehensive integrated genetic map of the complete karyotype of *Solea senegalensis* (Kaup 1858). *Genes*, 12, 49. https://doi.org/10.3390/genes12010049

Portela-Bens, S., Merlo, M., Rodríguez, M., Cross, I., Manchado, M., Kosyakova, N., Liehr, T., & Rebordinos, L. (2017). Integrated gene mapping and synteny studies give insights into the evolution of a sex proto-chromosome in *Solea senegalensis*. *Chromosoma*, 126, 261–277. https://doi.org/10.1007/s00412-016-0589-2

Rodríguez, M., Molina, B., Merlo, M., Arias-Pérez, A., Portela-Bens, S., García-Angulo, A., Cross, I., Liehr, T., & Rebordinos, L. (2019). Evolution of the proto sex-chromosome in *Solea senegalensis*. *International Journal of Molecular Sciences*, *20*, 5111. https://doi.org/10.3390/ijms20205111