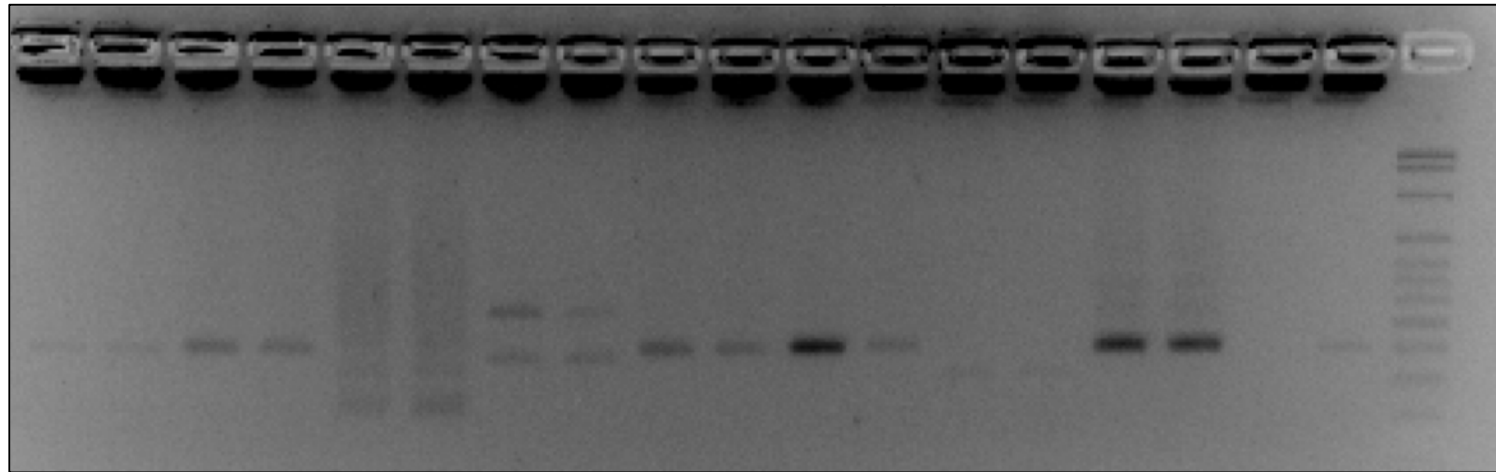


Supplementary Fig. S4. PCR-based validation of putative Z- and W-linked ddRAD markers in two-individual test panel of known-sex *Cryptobranchus* from Missouri. M = C36AM, F = C38AF, * = candidate retained for ten-individual panel.

(A) Candidate loci 1-9.

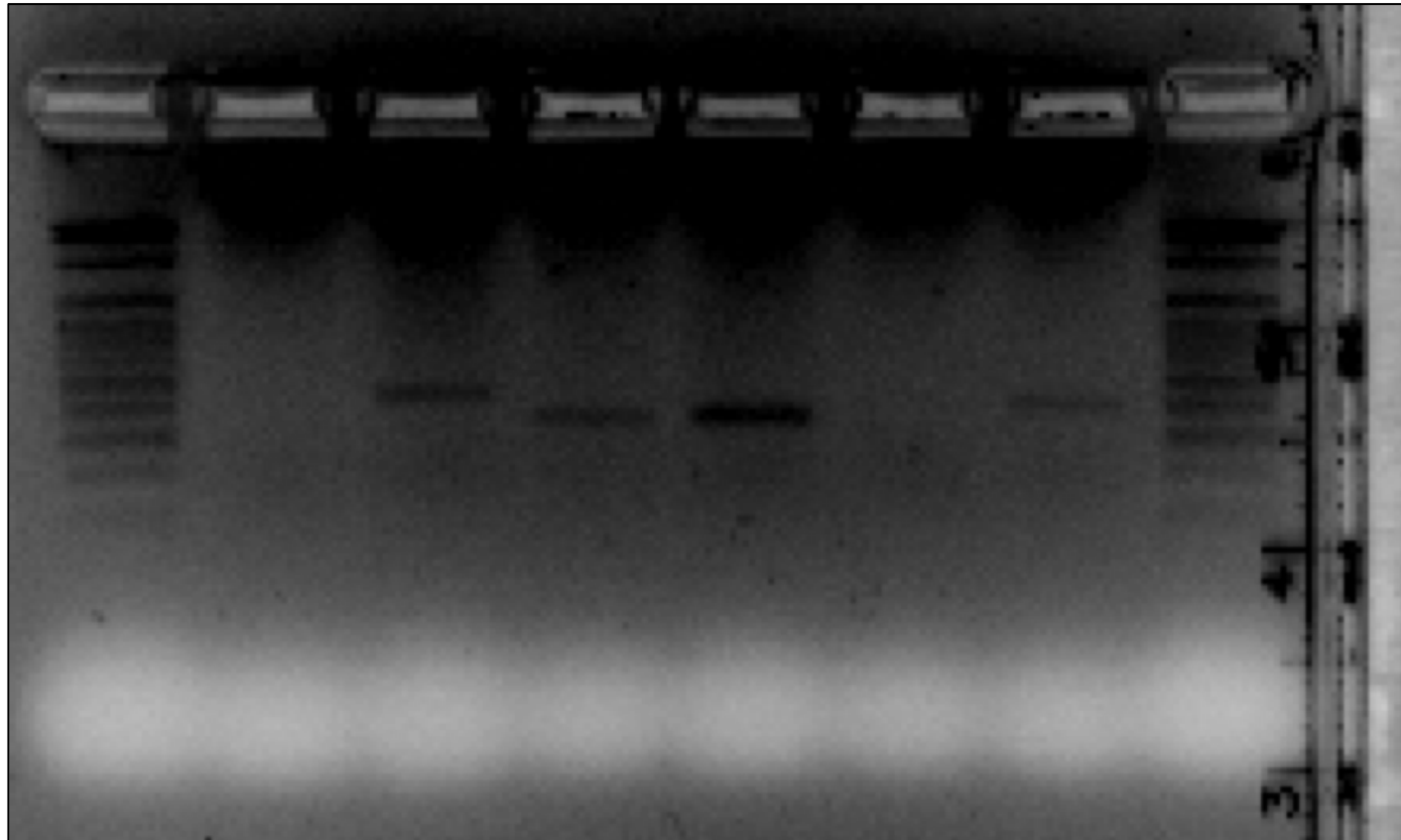
<u>Locus:</u>	1	1	2	2	3	3	4	4	5	5	6	6	7	7	8	8	9*	9*	Ladder
<u>Sex:</u>	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	



Supplementary Fig. S4. PCR-based validation of putative Z- and W-linked ddRAD markers in two-individual test panel of known-sex *Cryptobranchus* from Missouri. M = C36AM, F = C38AF, * = candidate retained for ten-individual panel.

(B) Candidate loci 10-12.

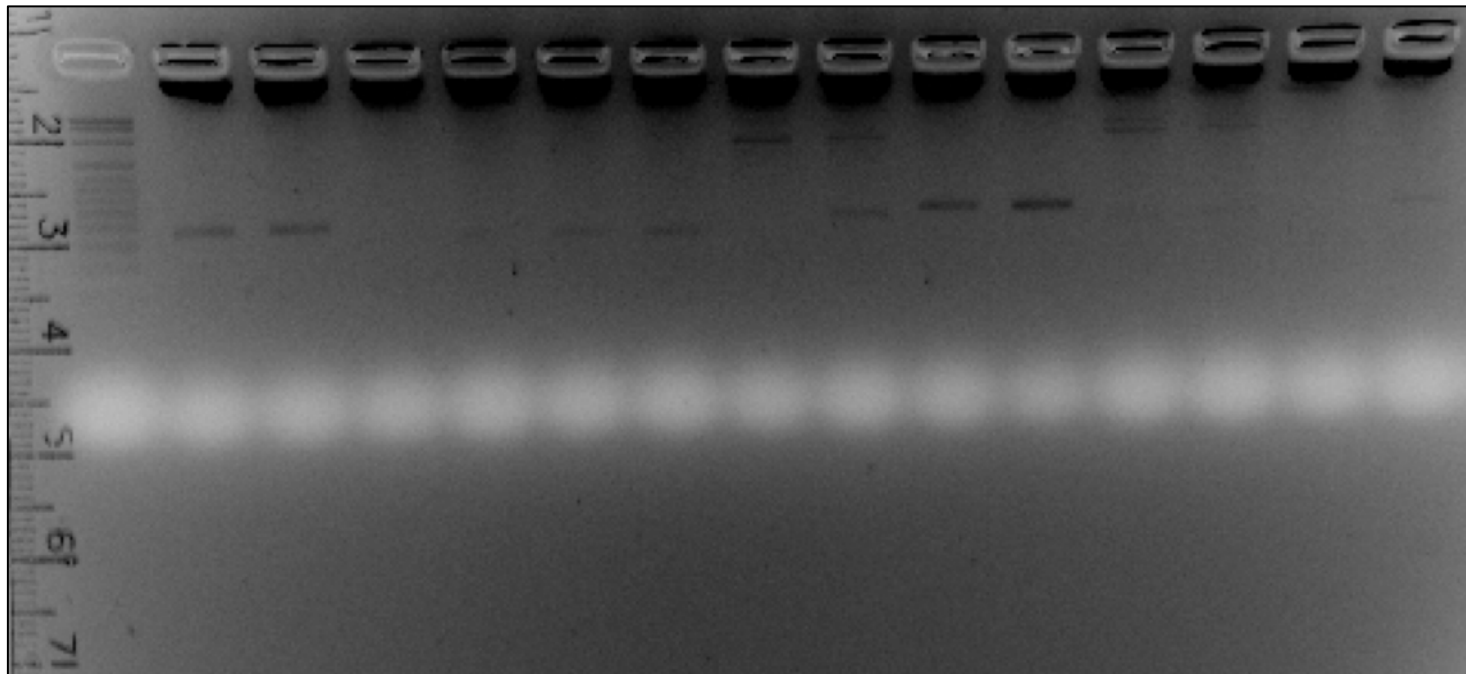
<u>Locus:</u>	10*	10*	11	11	12*	12*	Ladder
<u>Sex:</u>	M	F	M	F	M	F	



Supplementary Fig. S4. PCR-based validation of putative Z- and W-linked ddRAD markers in two-individual test panel of known-sex *Cryptobranchus* from Missouri. M = C36AM, F = C38AF, * = candidate retained for ten-individual panel.

(C) Candidate loci 13-19.

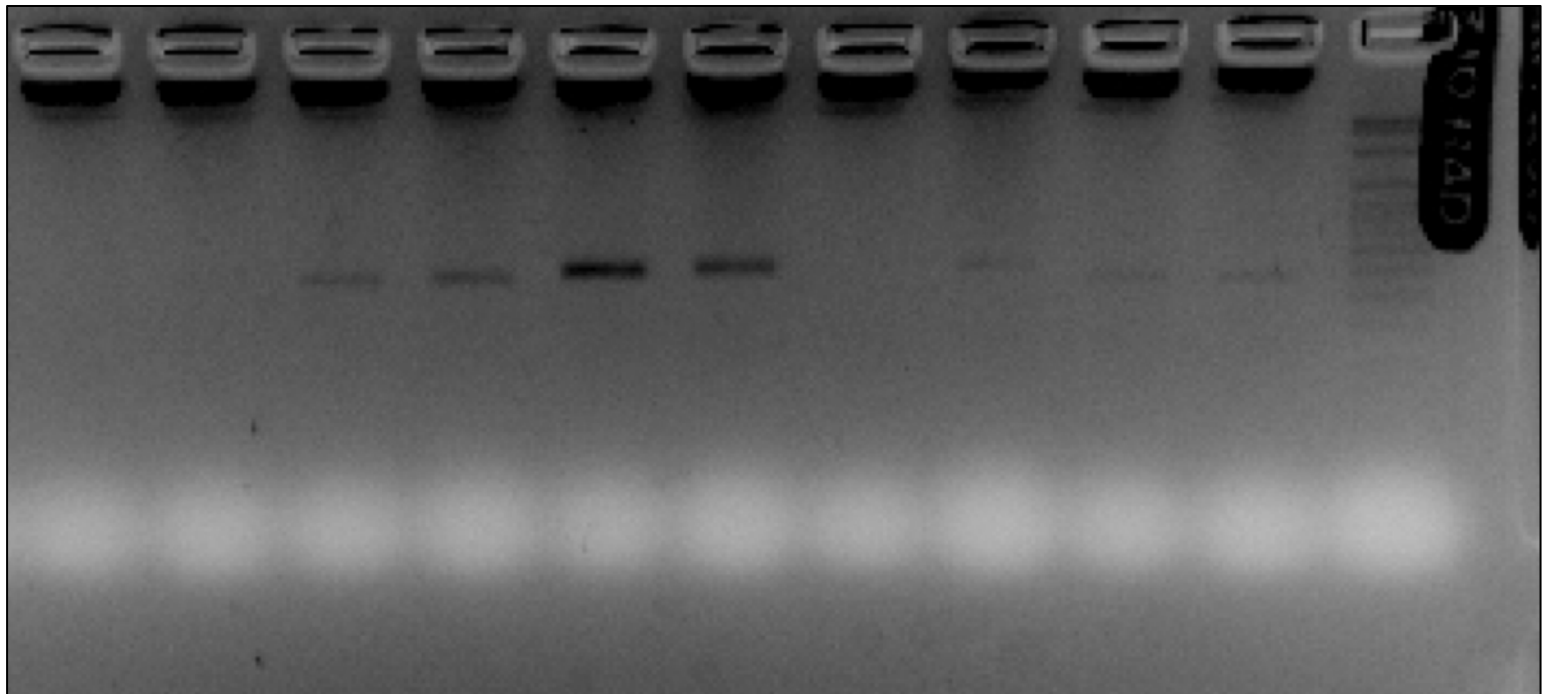
<u>Locus:</u>	Ladder	13	13	14*	14*	15	15	16*	16*	17	17	18	18	19*	19*
<u>Sex:</u>		M	F	M	F	M	F	M	F	M	F	M	F	M	F



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(D) Candidate loci 20-24.

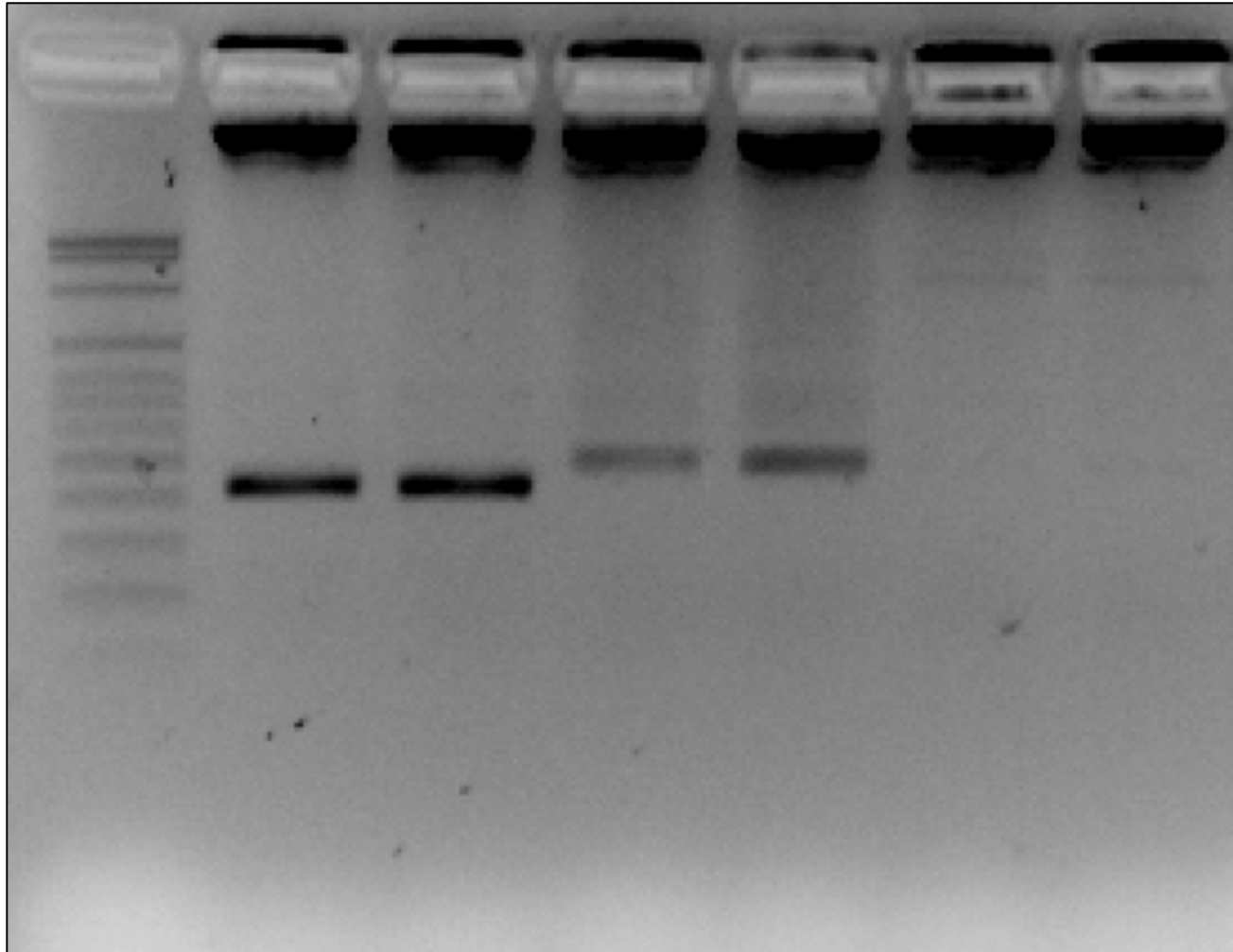
<u>Locus:</u>	20	20	21	21	22	22	23*	23*	24	24	Ladder
<u>Sex:</u>	M	F	M	F	M	F	M	F	M	F	



Supplementary Fig. S4. PCR-based validation of putative Z- and W-linked ddRAD markers in two-individual test panel of known-sex *Cryptobranchus* from Missouri. M = C36AM, F = C38AF, * = candidate retained for ten-individual panel.

(E) Candidate loci 25-27.

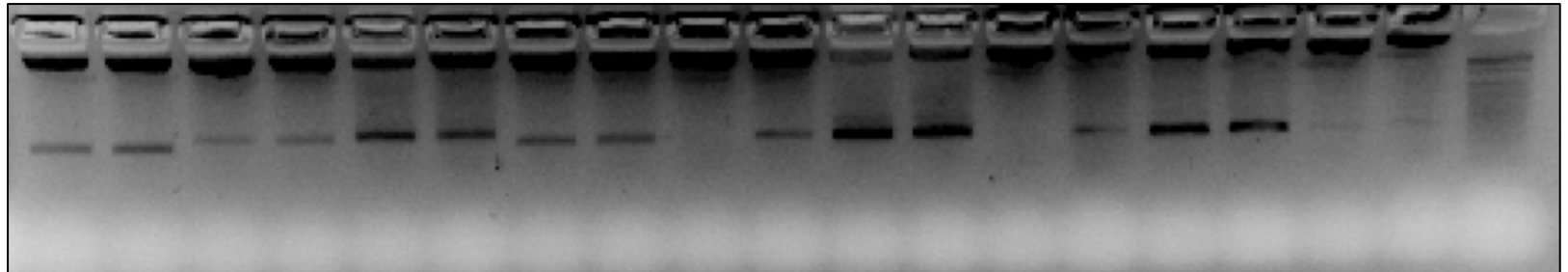
<u>Locus:</u>	Ladder	25	25	26	26	27	27
<u>Sex:</u>		M	F	M	F	M	F



Supplementary Fig. S4. PCR-based validation of putative Z- and W-linked ddRAD markers in two-individual test panel of known-sex *Cryptobranchus* from Missouri. M = C36AM, F = C38AF, * = candidate retained for ten-individual panel.

(F) Candidate loci 28-36.

<u>Locus:</u>	28	28	29	29	30	30	31	31	32*	32*	33	33	34*	34*	35	35	36	36	Ladder
<u>Sex:</u>	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	



Supplementary Fig. S4. PCR-based validation of putative Z- and W-linked ddRAD markers in two-individual test panel of known-sex *Cryptobranchus* from Missouri. M = C36AM, F = C38AF, * = candidate retained for ten-individual panel.

(G) Candidate loci 37-43 and 18S positive control.

<u>Locus:</u>	Ladder	37	37	38*	38*	39*	39*	40	40	41	41	42	42	43*	43*	N/A	N/A	18S	18S	Ladder
<u>Sex:</u>	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F		

