GENETIC MONITORING AND BIOLOGICAL CONTROL OF RECRUITMENT IN BONYTAIL REARING PONDS

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Southwestern Native ARRC

Dexter, NM

BACKGROUND

- The Southwestern Native ARRC in Dexter, NM maintains the only broodstock of *Gila elegans* (bonytail)
- Broodstock are spawned each year with larvae being reared at several facilities for grow-out
- The target stocking size requires a multi-year grow-out period and individuals can spawn during the second summer in grow-out ponds
- This inadvertent spawning may negatively impact growth and survival due to overcrowding in ponds
    - Poor water quality and oxygen depletions
- Genetic diversity may also be decreased as few individuals may contribute to offspring

OBJECTIVES

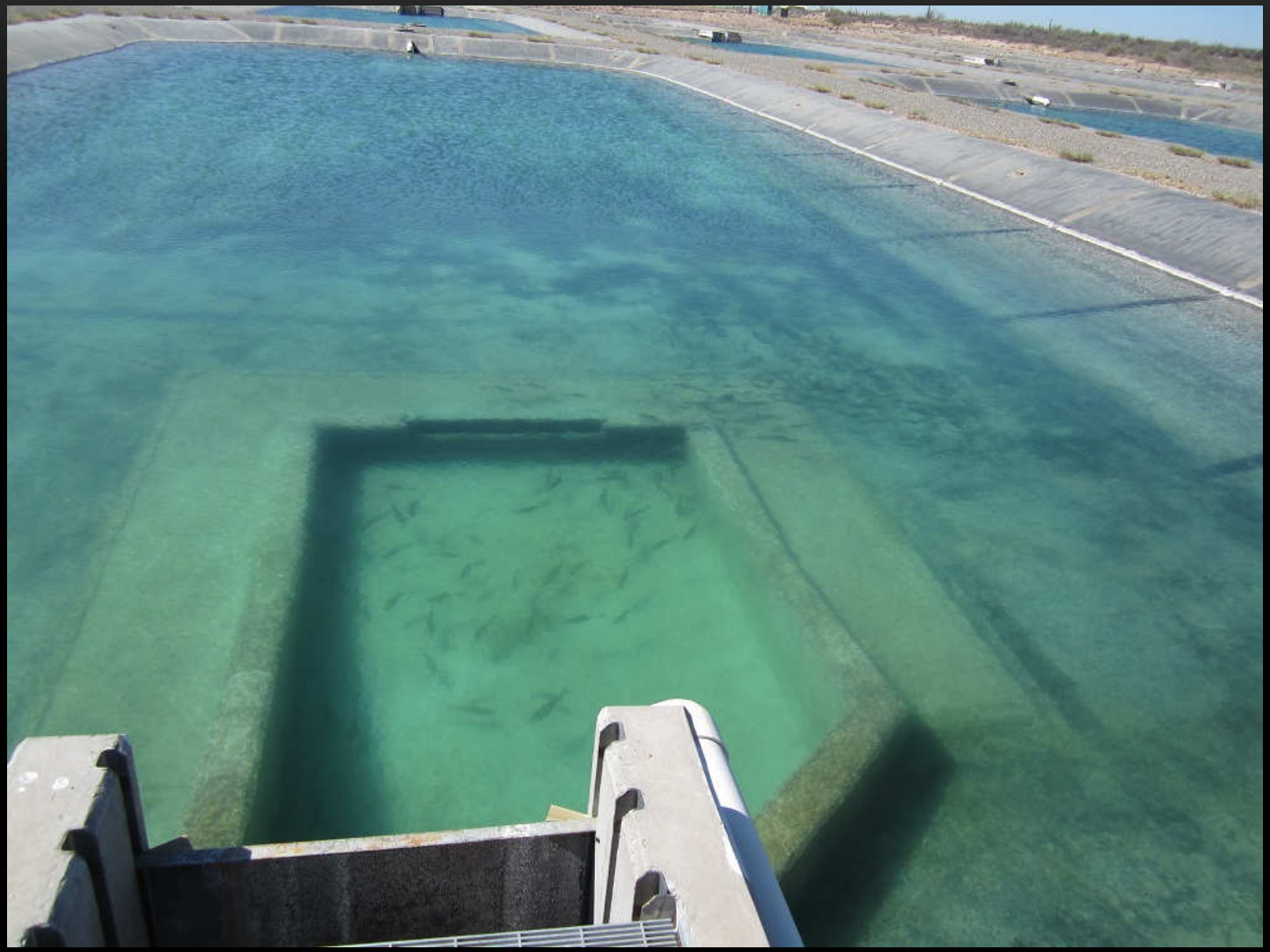
- To address the management implication of natural recruitment in grow-out ponds this study has two objectives:
  - Explore the use of Colorado pikeminnow as biological control and reduce recruitment in grow-out ponds
  - Use microsatellite markers to assess the genetic diversity and parentage of the offspring (recruits)

METHODS – YEAR ONE

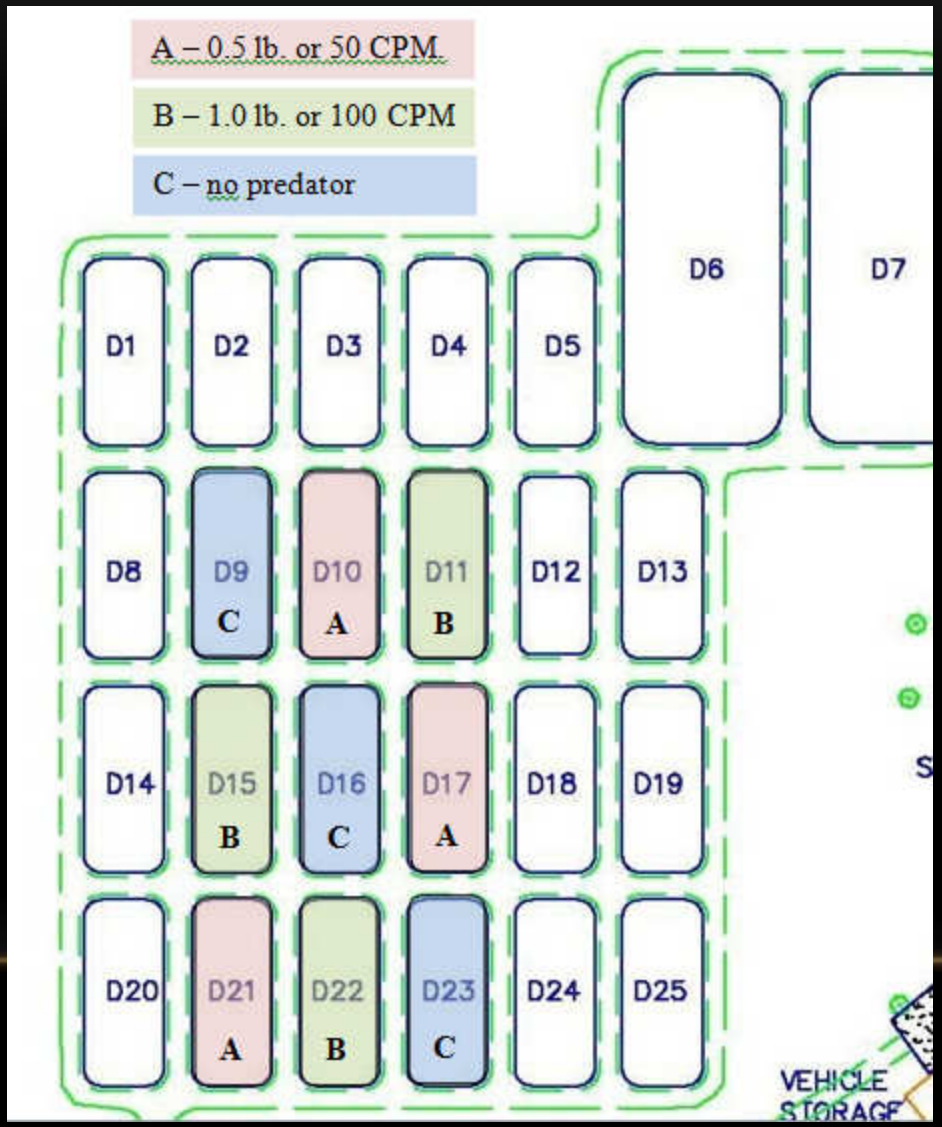
- Experimental Fish
  - Age-4 bonytail (2009 year class, mean total length 245.9 mm)
  - Age-0 Colorado pikeminnow (2012 year class, mean total length 60.6 mm)
  - All parental bonytail received PIT tags
- Experimental Design
  - 0.04 ha lined ponds
  - Three stocking treatments (three replicates per treatment)
    - Control – 100 male and 100 female bonytail, 0 Colorado pikeminnow
    - 50 CPM – 100 male and 100 female bonytail, 50 Colorado pikeminnow
    - 100 CPM - 100 male and 100 female bonytail, 100 Colorado pikeminnow
  - Ponds stocked in April and harvested in September (5 months)

METHODS – YEAR ONE

- Experimental Design (con't)
  - Initial lengths and weights were recorded on subsample of 20 bonytail and 10 Colorado pikeminnow
  - Water quality (temperature, DO)
  - Total number of offspring were calculated based on bulk weight
- Genetics
  - 20 microsatellite loci
  - Genotyped all adult bonytail (control ponds) & subsample of 100 offspring per pond
  - Descriptive statistics
    - Number of alleles
    - Allele frequency
    - Heterozygosity
  - Parentage Analyses – examine how many individuals contributed to offspring



EXPERIMENTAL POND LAYOUT



RESULTS

- Water quality was within acceptable ranges for the facility
- Dissolved oxygen ranged from 6.0 to 10.0 mg/L (a.m. readings)
- Temperature ranged from 12 ° C to 30 ° C
- Adult mortality was 1.8% with no difference among treatments
- Recruitment in the control ponds (0 Colorado pikeminnow) was significantly higher than ponds with 50 or 100 Colorado pikeminnow

NUMBER OF RECRUITS BY POND

		Number of individuals recruited	
Treatment	Pond #	By pond	By treatment
0 CPM	10D	3931	3752 (± 475.5)
	17D	4472	
	21D	2854	
50 CPM	11D	14	24 (± 6.1)
	15D	24	
	22D	35	
100 CPM	9D	62	25 (± 18.5)
	16D	11	
	23D	3	

GENETIC RESULTS - PRELIMINARY

- Two of the 20 loci were out of Hardy-Weinberg Equilibrium and removed for final analyses
- The total number of alleles did not differ
  - Among ponds
  - Between males, females or offspring within pond
- Heterozygosity was also similar with low inbreeding
- Allele frequencies were similar across ponds, genders, and parent-offspring
  - Even when frequency in parents was low
- Parentage analysis showed that many of the adults in each pond contributed
  - Some individuals contributed to multiple offspring
  - When females had multiple offspring many of the males were different

GENETIC SUMMARY STATISTICS

Control Pond	Number of Alleles	Observed Heterozygosity	Expected Heterozygosity	Inbreeding (F)
10D Females	10.632	0.776	0.812	0.043
10D Males	10.579	0.771	0.800	0.033
10D Offspring	10.158	0.765	0.795	0.033
17D Females	10.368	0.758	0.812	0.064
17D Males	10.684	0.771	0.785	0.010
17D Offspring	10.368	0.770	0.794	0.033
21D Females	10.368	0.768	0.807	0.052
21D Males	10.579	0.741	0.777	0.047
21D Offspring	10.474	0.771	0.804	0.042

ALLELE FREQUENCY BY POND

Locus	Allele	10DF	10DM	10DO	17DF	17DM	17DO	21DF	21DM	21DO
Gbig39	221	0.204	0.232	0.270	0.203	0.220	0.170	0.201	0.293	0.263
	223	0.102	0.131	0.085	0.135	0.125	0.170	0.098	0.157	0.131
	225	0.026	0.025	0.035	0.042	0.015	0.030	0.031	0.015	0.010
	231	0.020	0.010	0.025	0.010	0.005		0.005	0.020	0.005
	234	0.020	0.020	0.010	0.016	0.030	0.030	0.041	0.005	0.025
	238	0.036	0.056	0.050	0.031	0.035	0.045	0.041	0.030	0.005
	244	0.087	0.071	0.020	0.057	0.055	0.070	0.077	0.071	0.101
	250	0.031	0.015	0.015	0.016	0.020	0.015	0.010	0.015	0.025
	254	0.097	0.106	0.080	0.099	0.075	0.095	0.144	0.091	0.146
	255	0.066	0.051	0.090	0.089	0.095	0.105	0.041	0.081	0.051
	261	0.087	0.076	0.070	0.083	0.080	0.055	0.093	0.045	0.081
	267	0.133	0.152	0.160	0.177	0.190	0.160	0.170	0.116	0.116
	271	0.005								
	277	0.087	0.056	0.090	0.042	0.055	0.055	0.046	0.061	0.040

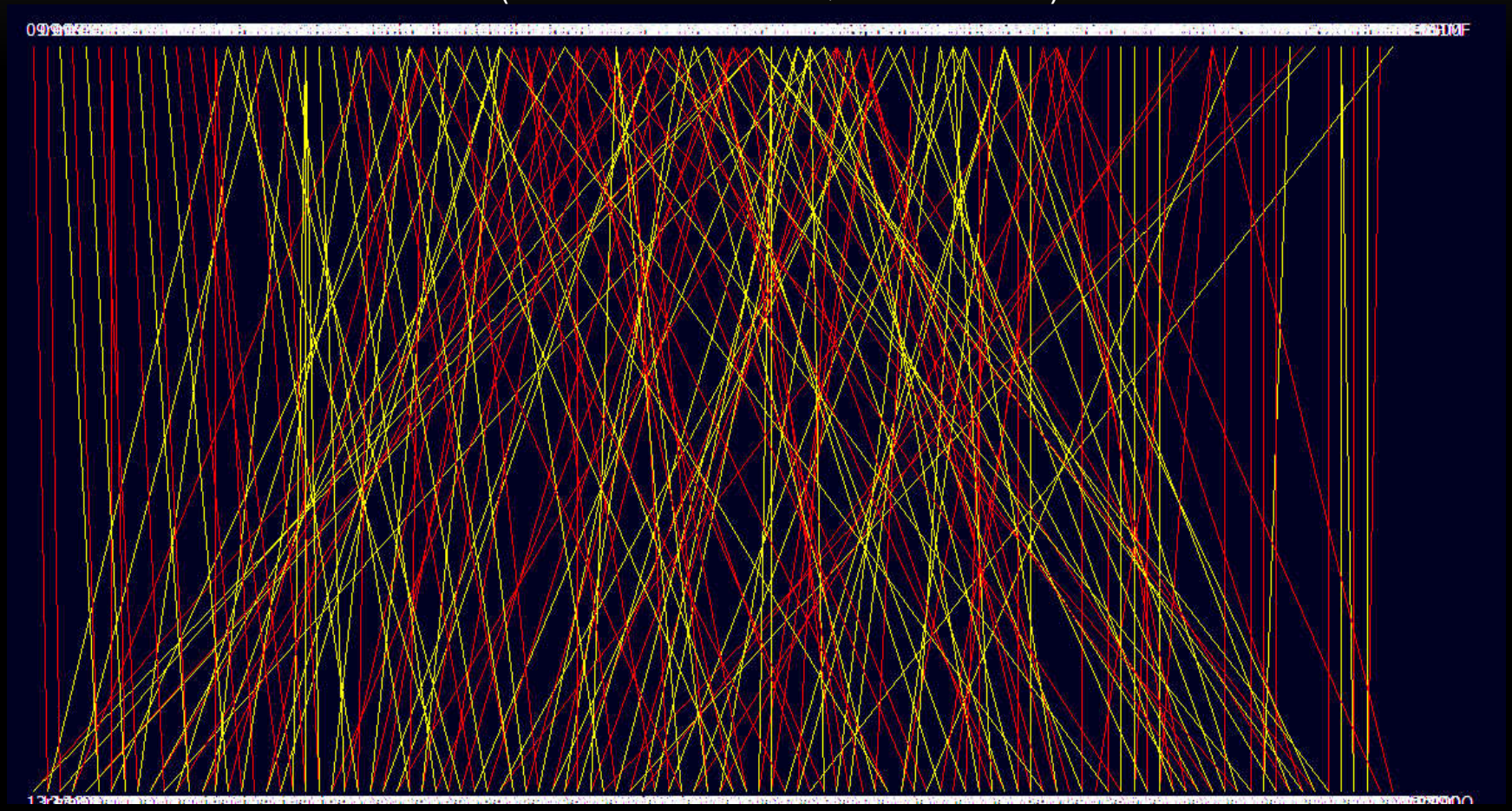
PARENT-OFFSPRING PEDIGREE – 1

PARENTS (YELLOW = FEMALE; RED = MALE)

OFFSPRING

PARENT-OFFSPRING PEDIGREE – 1

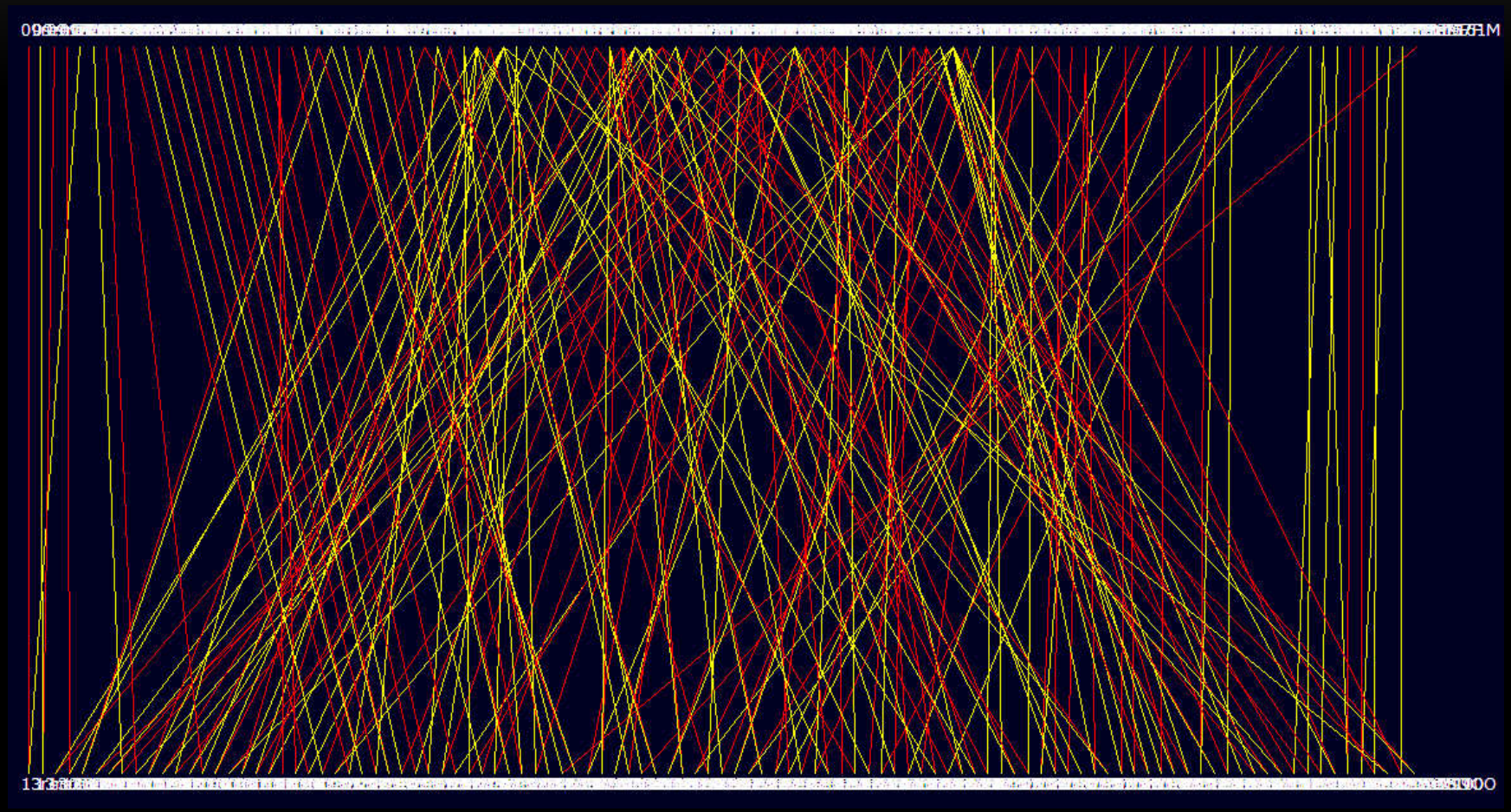
PARENTS (YELLOW = FEMALE; RED = MALE)



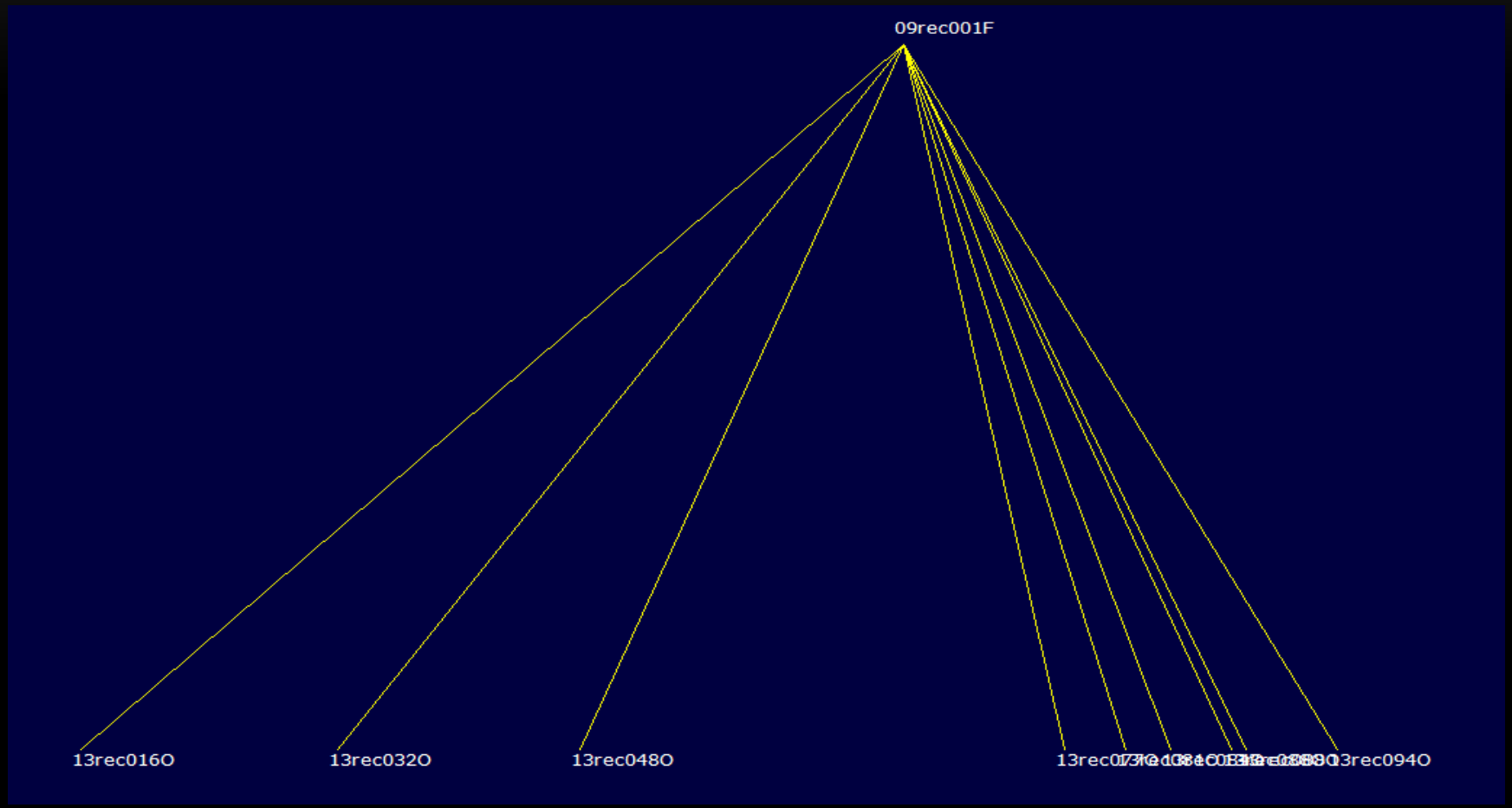
OFFSPRING

PARENT-OFFSPRING PEDIGREE - 2

PARENT-OFFSPRING PEDIGREE - 2



PARENT-OFFSPRING PEDIGREE - 2



CONCLUSIONS

- Colorado pikeminnow controls natural recruitment even when using 50 CPM
- Initially is seems that genetic diversity of offspring is similar to adults with little inbreeding
- Some adults contributed to multiple offspring, but offspring were often half-sibs
  - Offspring having same female parent had different male parents
    - If single large clutch then there is random egg fertilization by various males due to sperm competition
    - Some females may also produce smaller clutches allowing different males to contribute
    - Did not genotype every offspring individual, so complete picture of adult contributions is unknown
- Currently analyzing year two results: stocked in May and harvested in October

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THANK YOU!