

Genomic Background and Generation Time Influence Deleterious Mutation Rates in *Daphnia*

Leigh C. Latta IV,^{*1} Kendall K. Morgan,[†] Casse S. Weaver,[‡] Desiree Allen,[§] Sarah Schaack,^{*} and Michael Lynch^{**}

^{*}Department of Biology, Reed College, Portland, Oregon 97202, [†]Institute for Genome Sciences and Policy, Duke University, Durham, North Carolina 27708, [‡]Eureka, California 95503, [§]Centre for Evolutionary Biology, University of Western Australia, Perth, Western Australia 6009, Australia, and ^{**}Department of Biology, Indiana University, Bloomington, Indiana 47405

ABSTRACT Understanding how genetic variation is generated and how selection shapes mutation rates over evolutionary time requires knowledge of the factors influencing mutation and its effects on quantitative traits. We explore the impact of two factors, genomic background and generation time, on deleterious mutation in *Daphnia pulex*, a cyclically parthenogenic aquatic microcrustacean, using parallel mutation-accumulation experiments. The deleterious mutational properties of life-history characters for individuals from two different populations, and for individuals maintained at two different generation times, were quantified and compared. Mutational properties varied between populations, especially for clutch size, suggesting that genomic background influences mutational properties for some characters. Generation time was found to have a greater effect on mutational properties, with higher per-generation deleterious mutation rates in lines with longer generation times. These results suggest that differences in genetic architecture among populations and species may be explained in part by demographic features that significantly influence generation time and therefore the rate of mutation.

AS the ultimate source of all genetic variation, mutation is an important evolutionary force affecting the ability of natural populations to respond to selective pressures. Most spontaneous mutations are deleterious (Lynch *et al.* 1999; Eyre-Walker and Keightley 2007), which is thought to explain many evolutionary phenomena, including inbreeding depression, mating system evolution, senescence, and risk of extinction to small populations (Charlesworth and Charlesworth 1998; Lynch *et al.* 1999). Despite the importance of knowing mutation rates in both theoretical and applied biology, few empirical estimates exist other than those for classic genetic model organisms (Baer *et al.* 2007), and little is known about the factors influencing the rate of mutation among individuals, populations, and species (Lynch 2010).

In addition to direct estimates based on sequencing, estimates of the parameters for mutations affecting fitness

[*i.e.*, the genome-wide deleterious mutation rate (U) and the average effect (\bar{s})] have now been reported for several species (reviewed in Baer *et al.* 2007). However, little empirical attention has been given to variability in the phenotypic effects of deleterious mutation [*i.e.*, per-generation rates of change in the mean phenotype (ΔM) and mutational variance (ΔV)] or to the associated deleterious mutation parameters that can be inferred from these quantities (U and \bar{s}) among populations within a species. Recent theoretical treatments of mutation-rate evolution, however, predict individual variation in mutation rates (Lynch 2008; Desai and Fisher 2011) and fitness dependence of mutation rates (Agrawal 2002; Shaw and Baer 2011), highlighting the importance of this variability.

The deepest understanding comes from recent mutation-accumulation studies in *Drosophila melanogaster* that provide evidence for variability in mutation rates among genotypes, using both direct methods of rate estimation based on sequence data (Haag-Liautard *et al.* 2007) and indirect methods using fitness data (Avila *et al.* 2006). Interestingly, intraspecific variability in mutation rates in *Drosophila* appears to be correlated with the quality of the genomic background in which mutations accumulate: genotypes of

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¹Corresponding author: Department of Biology, Reed College, 3203 S.E. Woodstock Blvd., Biology Bldg., Room 104, Portland, OR 97202. E-mail: llatta@reed.edu

poor condition (harboring a high number of deleterious mutations initially) display higher mutation rates than genotypes with fewer starting mutations (Avila *et al.* 2006; Agrawal and Wang 2008; Sharp and Agrawal 2012). In contrast to the situation in *Drosophila*, where both direct and indirect estimates suggest intraspecific mutation-rate variability, in rhabditid nematodes indirect estimates of mutation rate (Baer *et al.* 2005) and direct estimates of insertion–deletion mutation processes (Phillips *et al.* 2009) suggest variability among genotypes, but direct estimates of the base-substitution mutation rate in the same genotypes imply rate homogeneity (Denver *et al.* 2012).

If mutation rate variability is fitness dependent, as appears to be the case in *Drosophila*, it suggests that life-history traits facilitating the production of higher deleterious mutation loads could drive evolution of the mutation rate. Generation time, in particular, is a life-history trait that influences mutation load, given that the number of mutations that an individual inherits depends on the number of parental germline cell divisions and germline divisions increase with generation time (reviewed in Bronham 2009). Thus, populations with short generation times are expected to undergo fewer germline cell divisions than populations with long generation times, thereby passing on fewer mutations to their offspring. The numerous examples of male mutation bias, whereby males undergo more germline cell divisions and generate more mutations each generation than females, provide compelling evidence for such generation-time effects (Sayres and Makova 2011; Kong *et al.* 2012).

In this study, we compare the patterns of deleterious mutation accumulation in individuals from two natural populations of *Daphnia pulicaria* from Oregon. These populations were chosen based on their divergent ecological origins. One population, Klamath Lake, is from a lake in the southern Cascade Mountains, while the other population, Lake Marie, is located near the Pacific coast. Specifically, these populations represent the high and low extremes of the phenotypic distribution for life-history traits and body size of natural populations in Oregon (Morgan *et al.* 2001; Baer and Lynch 2003). Not only do phylogenetic analyses identify these populations as among the most genetically divergent in western Oregon, but also quantitative assessments of the expressed levels of genetic variation show that Lake Marie *D. pulicaria* are low relative to Klamath Lake *D. pulicaria*, despite similar levels of genetic diversity for both allozyme and microsatellite loci in these populations (Morgan *et al.* 2001). In addition to the population comparison, for one population (Klamath Lake) we experimentally imposed different generation times to assess the impact on the mutation parameters. By exploring the properties of spontaneous deleterious mutation in multiple genotypes from two divergent populations, as well as by manipulating generation time in one population, we characterize the variability in deleterious mutation properties that arises due to genomic background and assess whether differences in generation time provide an explanation for such variability.

Methods

Mutation accumulation experiment

Ten *D. pulicaria* individuals (genotypes) from each of two Oregon lakes, Klamath Lake and Lake Marie (populations), were isolated into individual beakers. Each founding individual was allowed to reproduce clonally until 10 genetically identical offspring were produced (clones). These 10 replicate clones of each genotype from each population were then placed into their own beakers to establish 200 mutation-accumulation (MA) lines for the subsequent experiments (two populations \times 10 genotypes \times 10 clones = 200 MA lines). All MA lines were maintained under constant environmental conditions [at 18°, 12 light (L):12 dark (D) light cycle] in a controlled environmental chamber.

Mutation-accumulation lines were propagated by single-offspring descent by haphazardly choosing a juvenile every generation (see below for conditions for short and long generation times). Parental lines were kept as backups until the next transfer was performed to allow for the replacement of lost lines. In this manner, the 10 clonal lines started from each genotype were allowed to diverge based on incoming mutations. In contrast to the MA lines, control lines of each genotype were maintained under conditions designed to greatly slow the rate of deleterious mutation accumulation (at 7°, 12L:12D light cycle, in large populations, without single-offspring descent).

For both populations, MA lines were propagated by random offspring from the second clutch [short generation time (SGT) lines]. For one population (Klamath Lake), a parallel set of long generation time (LGT) lines was constructed, where the propagating juvenile was obtained from the last possible clutch. In this way, all three sets of lines (Lake Marie_{SGT}, Klamath_{SGT}, and Klamath_{LGT}) were maintained under identical conditions (at 18°, 12L:12D light cycle), except that the Klamath_{LGT} lines were manipulated to have a longer generation time. At the close of the experiment, the Lake Marie_{SGT} and Klamath_{SGT} lines had undergone 65 generations of divergence on average, while the Klamath_{LGT} lines had undergone only 25 generations of divergence over the same absolute period of time. Thus, the generation time was extended by a factor of 2.5 in the Klamath_{LGT} lines.

Phenotypic assays

Representatives of each surviving MA line, along with controls, were phenotypically assayed using a standard life table design (Lynch 1985). Assays were conducted after Lake Marie_{SGT} and Klamath_{SGT} lines had diverged for ~15, 30, and 65 generations. Measurements from phenotypic assays were used to calculate the mean and variance for life-history characters among diverging lines within and among genotypes from each population. Prior to each assay, each line was replicated into three sublines, each of which was then taken through two generations of clonal reproduction under controlled conditions (18°, 12L:12D light cycle).

Such an experimental design ensures that maternal and grandmaternal effects contribute to the environmental (within-line) rather than the genetic (among-line) component of variance in the final analysis (Lynch 1985). During the life-history assays, the following measurements were made: time to maturity, size at maturity (to the nearest 0.01 mm), and clutch size in each of the first four clutches.

Data analysis

For each of the surviving sets of lines in each of the assays, the within- and among-line components of variance were extracted by one-way analysis of variance. The within-line variance represents the environmental variance (V_E) and the among-line variance represents the genetic variance (V_G). Estimates of the rate of change in the genetic variance were obtained by weighted least-squares regression of the V_G estimates on generation number, with the slope of the regression representing an estimate of the rate of change in the variance due to mutational input, ΔV (Lynch and Walsh 1998). Data points in the regression were weighted by the inverse of the sampling variance of V_G . The final estimates of ΔV for Lake Marie_{SGT}, Klamath_{SGT}, and Klamath_{LGT} lines were obtained by averaging the line-specific estimates. The standard error of this estimate was obtained by treating the line-specific estimates as independent as described in Lynch *et al.* (1998). Estimates of the rate of change in the mean due to mutation accumulation, ΔM , were obtained by weighted least-squares regression of the assay-specific phenotypic means on generation number. Due to line extinction over the course of the mutation-accumulation phase in the Lake Marie_{SGT} lines, estimates of ΔV and ΔM were calculated using data obtained in all three phenotypic assays in some cases ($n = 2$) and from two phenotypic assays when necessary ($n = 5$).

For each assay, 10 control representatives of a subset of the founding clones (three Klamath clones and two Marie clones) were isolated from the larger, low-temperature (7°) control populations (each of which was then replicated into three sub-lines, as in the case of the MA lines). Measurements from control lines were used to calculate a mean and variance (among the 10 isolated individuals) for each founding clone at each assay. Regression analyses of mean control phenotype and among-line variance were then conducted such that estimates of ΔM and ΔV for MA lines could be corrected for change due to environmental variation among assays. Lower-bound estimates of the genomic mutation rate, $U_{min} = \Delta M^2 / \Delta V$, and upper-bound estimates of the heterozygous mutational effect as a fraction of the initial mean phenotype (\check{z}_0), $a'_{max} = \Delta V / \Delta M / \check{z}_0$, were obtained according to the method of Bateman (1959) and Mukai *et al.* (1972) and modified for clonal lines (Lynch and Walsh 1998). Bateman–Mukai estimates were corrected for bias caused by sampling error in estimating ΔM and ΔV (Lynch 1994). Standard errors for U_{min} and a'_{max} were obtained using the formula for the standard error of a ratio, taking the sampling variances of the numerator and denominator into account (Lynch and Walsh 1998).

Table 1 Per-generation rates of change in control mean phenotype, and among-line variance, over the three life-history assays

Trait	Mean	Variance
Size at maturity (mm)	0.0001 (0.0066)	0.0000 (0.0000)
Clutch size	−0.0536 (0.1000)	−0.0179 (0.1100)
Age at maturity (days)	0.0099 (0.0059)	−0.0089 (0.0100)

Reported values are coefficients for regressions on generation number; standard errors are given in parentheses. Estimates for clutch size represent an average value of the separate analyses of clutches 1–4. None of the estimates deviates significantly from zero. The variance for size at maturity was $<10^{-4}$.

It should be noted that application of the Bateman–Mukai technique, which assumes equal, unidirectional mutational effects, is unlikely to generate realistic estimates of deleterious mutation parameters (Halligan and Keightley 2009). Lake-dwelling *Daphnia* populations, including those used here, are exposed to strong directional selection via size-selective vertebrate predation (*e.g.*, Gliwicz and Boavida 1996), which may, however, justify the assumption of unidirectional mutational effects. Furthermore, estimates obtained are specific to the assay environment employed, given the environmental dependence of spontaneous mutations (Kondrashov and Houle 1994; Fry and Heinsohn 2002). Despite these limitations, the method is informative in that it: (1) provides lower- and upper-bound estimates of deleterious mutation parameters and (2) can be used to assess the impact of genomic background and generation time within a given experiment.

Results

Mutation accumulation in divergent populations

The phenotypic means and variances of the control lines did not vary significantly over the course of the experiment (Table 1; Supporting Information, Table S1). Therefore, the estimates of ΔM and ΔV from the mutation-accumulation lines were not corrected for environmental variation among assays, and we interpret changes in the MA lines to be a consequence of genetic, rather than environmental, change.

The estimates of ΔM are identical between the Klamath_{SGT} and Lake Marie_{SGT} lines for size at maturity, with an increase in body size in response to new mutations (Table 2; Table S2). However, the two populations differ by an order of magnitude in the rate of decline in mean clutch size, with the Lake Marie_{SGT} lines experiencing an elevated rate relative to the Klamath_{SGT} lines. Klamath_{SGT} lines also showed a significant rate increase in age at maturity relative to the Lake Marie_{SGT} lines. Estimates of ΔV for the Klamath_{SGT} lines are quite consistent with those for Lake Marie_{SGT} lines, with none of the values differing significantly (Table 2; Table S3).

Application of the observed temporal changes in the mean and the genetic variance to the Bateman–Mukai estimators yields lower-bound estimates of the mutation rate (U_{min}) for body size that are nearly identical for the Klamath_{SGT} and Lake Marie_{SGT} lines (Table 2; Table S4). In contrast, the

Table 2 Estimates of the initial mean phenotype (\bar{z}_0), per-generation rate of change in the mean phenotype (ΔM), per-generation rate of input of mutational variance (ΔV), genomic mutation rate (U_{min}), and average mutational effect as a fraction of the initial mean phenotype (a'_{max}) for Lake Marie, Klamath_{SGT}, and Klamath_{LGT} lines

Trait	Population	\bar{z}_0	ΔM	ΔV	U_{min}	a'_{max}
SM	Marie	1.84 (0.02)	0.003 (0.001)	0.0001 (0.0000)	0.027 (0.001)	0.013 (0.001)
	Klamath _{SGT}	2.03 (0.03)	0.003 (0.001) ^a	0.0001 (0.0000) ^a	0.028 (0.001) ^a	0.012 (0.001) ^a
	Klamath _{LGT}	2.03 (0.03)	0.012 (0.002)	0.0002 (0.0001)	0.397 (0.069)	0.006 (0.000)
CS	Marie	7.63 (0.45)	-0.194 (0.055)	0.1299 (0.0930)	0.157 (0.014)	-0.082 (0.031)
	Klamath _{SGT}	8.11 (1.01)	-0.020 (0.018) ^{a,b}	0.1114 (0.0380) ^a	0.002 (0.000) ^{a,b}	-0.141 (0.055) ^a
	Klamath _{LGT}	8.11 (1.01)	-0.061 (0.059)	0.2216 (0.1048)	0.015 (0.046)	-0.022 (0.000)
AM	Marie	6.61 (0.28)	-0.002 (0.005)	0.0088 (0.0030)	NA	NA
	Klamath _{SGT}	6.38 (0.30)	0.007 (0.002) ^{a,b}	0.0038 (0.0020) ^a	0.006 (0.000) ^a	0.074 (0.012)
	Klamath _{LGT}	6.38 (0.30)	0.019 (0.012)	0.0102 (0.0020)	0.010 (0.000)	0.060 (0.010)

Estimates for clutch size represent an average value of the separate analyses of clutches 1–4. Standard errors are given in parentheses. SM, size at maturity (mm); CS, clutch size; and AM, age at maturity (days). Estimates of U_{min} and a'_{max} were not calculated for Marie AM because the standard error of ΔM exceeds the mean (indicated as NA).

^a Indicates a significant difference between Klamath_{SGT} and Klamath_{LGT} lines.

^b Indicates a significant difference between Marie and Klamath *D. pulicaria* populations.

mutation rate for clutch size in Lake Marie_{SGT} lines is significantly higher than the rate estimated for the Klamath_{SGT} lines. Upper-bound estimates of mutational effects as a fraction of the initial mean phenotype (a'_{max}) are all <15% of the initial mean phenotypic value and do not vary significantly between populations.

Generation-time effect on mutation accumulation

Estimates of ΔM for the Klamath_{LGT} lines are on average 3.3 times greater than those for the Klamath_{SGT} lines (Table 2; Table S2). Similarly, estimates of ΔV for the Klamath_{LGT} lines are on average 2.2 times greater than those for the Klamath_{SGT} lines, which is very similar to the 2.5-fold difference in generation time between the two sets of lines (Table 2; Table S3). For each of the traits measured, the per-character estimate of U_{min} for the Klamath_{LGT} lines significantly exceeds that for the Klamath_{SGT} lines (Table 2). The estimates of a'_{max} are significantly elevated in the Klamath_{SGT} lines relative to the Klamath_{LGT} lines for both body size and clutch size, but not for age at maturity (Table 2).

Discussion

Our data demonstrate trait-specific variation in mutation parameters among natural isolates from lake-dwelling populations of *D. pulicaria* that have diverged phenotypically and genetically through natural processes, a result consistent with other recent studies showing high levels of intraspecific variation in mutation rates among strains in classic model organisms (Baer *et al.* 2005; Haag-Liautard *et al.* 2007). Furthermore, we show evidence for a generation time effect on mutation rate estimates, providing insight into a possible mechanism explaining mutation rate variability among populations and species where ecological conditions or demographics may influence this feature of the life history.

Among the differences in mutation parameters observed between populations, the largest change was the rate of

decline in clutch size, which may be explained by several factors. First, low levels of expressed genetic variation observed in the Marie population, in conjunction with the high levels of molecular variation (Morgan *et al.* 2001), may indicate that this population maintains substantial levels of hidden quantitative genetic variation. Hidden genetic variation could result from prolonged periods of asexual reproduction leading to the accumulation of mutations (Barton and Charlesworth 1998). If the Marie population harbors an elevated base mutation load, this may lead to an increased rate of change in the phenotype because new mutations are predicted to accumulate faster in more loaded populations (Agrawal 2002; Shaw and Baer 2011). Given that the Marie lines experienced a greater rate of extinction over the course of the mutation accumulation experiment, an indication of higher mutation load (Lynch 1994), and exhibited large reductions in competitive ability (a complex fitness trait) as a result of mutation accumulation relative to Klamath Lake (Schaack *et al.* 2012), the possibility of high mutation load in this population is plausible.

Alternatively, the elevated rate of change in clutch size in Marie may reflect a difference in genotype × environment interactions between the populations. In the current study, all MA lines were assayed in the same benign laboratory environment, but this environment may represent different levels of stress for animals originating from the Marie vs. Klamath Lake populations (Baer and Lynch 2003). Decreased reproductive success in Marie may be due to magnification of mutational effects and/or increases in the number of mutations that produce a measurable effect due to stress, if the laboratory environment differs from the field environment to different degrees (Kondrashov and Houle 1994; Fry and Heinsohn 2002).

The magnitude of our trait-specific estimates of the rates of change in mean phenotype and the variance in *D. pulicaria* are generally lower than those obtained from MA experiments conducted under similar environmental conditions

involving the pond-dwelling sister species, *Daphnia pulex* (Lynch *et al.* 1998). These differences in rates of change in the mean and variance translate into lower estimates of the mutation rate and higher estimates of the average mutational effect in *D. pulicaria* relative to *D. pulex*. Also, while the direction of phenotypic change in response to new mutations is consistent across the two *D. pulicaria* populations, with body size increasing and clutch size decreasing, this directional change is opposite the results for *D. pulex* [decreased body size and increased clutch size (Lynch *et al.* 1998)]. This observation suggests that there is a characteristic direction of mutational effects among divergent populations within a species, but that the direction of effects among species may differ, perhaps resulting from differences in selection pressures in lakes vs. ponds (Dudycha and Tessier 1999).

The positive relationship between generation time and the estimate of per-generation mutation rate is consistent with previous data based on broad, indirect comparisons among organisms with a wide range of generation times (Lynch *et al.* 1999; Keightley and Eyre-Walker 2000). The close match between the differences that we observed and the experimentally manipulated 2.5-fold difference in generation time between treatments within a species, however, provides more direct evidence supporting the hypothesis that per-generation mutation rates scale with the absolute generation time by circumventing the confounding effects of an interspecific comparison.

An effect of generation time may be largely due to the increase in germline cell divisions occurring in older parents, which result in additional opportunities for DNA replication error (Li *et al.* 1996; Drake *et al.* 1998). In *Daphnia*, longevity varies among closely related populations and species according to habitat permanence [*e.g.*, permanent lakes vs. temporary ponds (Dudycha and Tessier 1999)]. Thus, environmental factors may shift the average maternal age in this genus, leading to the significant differences in per-generation mutation rates, levels of standing genetic variation, and mutation loads empirically predicted by ecological and evolutionary models (*e.g.*, Hansen and Price 1999).

Over long time periods, because generation time is inversely related to population size (Ohta and Kimura 1971; Ohta 1987), populations with long generation times may exhibit not only increased per-generation mutation rates, but also a reduced ability to purge deleterious mutations due to small effective population size (Lynch and Conery 2003). The resulting high deleterious mutation load may lead to a further increase in mutation rate (Avila *et al.* 2006; Sharp and Agrawal 2012), meaning that small populations with long generation times will exhibit accelerated declines in fitness and lower viability for multiple reasons. Thus, the intraspecific variation and generation-time effects reported here may be important considerations for future work on biological phenomena, in both the theoretical and applied realm, that depend on mutation rate estimates and their variability among populations and species.

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Genomic Background and Generation Time Influence Deleterious Mutation Rates in *Daphnia*

**Leigh C. Latta IV, Kendall K. Morgan, Casse S. Weaver, Desiree Allen, Sarah Schaack,
and Michael Lynch**

Table S1 Trait-specific means and two standard errors for control genotypes from each of the two populations (Klamath and Marie) for each of the three phenotypic assays.

Klamath						
SM	Mean 1	2SE1	Mean 2	2SE2	Mean 3	2SE3
G	2.23	0.06	2.42	0.06	2.25	0.05
I	2.05	0.08	2.36	0.04	2.15	0.06
K	1.83	0.06	2.32	0.06	1.95	0.06
CL1	Mean 1	2SE1	Mean 2	2SE2	Mean 3	2SE3
G	3.79	1.58	6.53	1.16	4.16	0.88
I	2.92	1.42	3.47	1.06	1.73	0.8
K	3.5	1.26	6.61	1.52	0.73	0.54
CL2	Mean 1	2SE1	Mean 2	2SE2	Mean 3	2SE3
G	7.46	3.42	19	2.44	10.23	1.72
I	6.83	1.76	8.87	2.46	4.79	1.52
K	8.76	1.88	13.67	2.12	1.33	0.98
CL3	Mean 1	2SE1	Mean 2	2SE2	Mean 3	2SE3
G	13.27	4.84	20.3	2.74	10.43	1.86
I	8.33	2.68	8.07	2.54	5.51	1.98
K	10.61	3.24	18.11	2.78	2.79	1.22
CL4	Mean 1	2SE1	Mean 2	2SE2	Mean 3	2SE3
G	15.9	4.38	24	4.48	11.17	2.72
I	8.92	3.72	11.63	3.76	7.51	2.12
K	13.46	2.5	21.72	3.58	2.48	1.58
AM	Mean 1	2SE1	Mean 2	2SE2	Mean 3	2SE3
G	6.07	0.66	6.17	0.42	6.91	0.4
I	6.76	0.6	6.06	0.3	6.65	0.46
K	6.02	0.5	6.34	0.5	6.6	0.26
Marie						
SM	Mean 1	2SE1	Mean 2	2SE2	Mean 3	2SE3
B	1.71	0.104	1.8	0.056	1.83	0.084
K	1.89	0.076	1.93	0.056	1.9	0.108
CL1	Mean 1	2SE1	Mean 2	2SE2	Mean 3	2SE3
B	3.54	1.476	0.87	0.92	1.1	1.26

K	4.93	1.48	4.7	2.12	1.39	0.96
CL2	Mean 1	2SE1	Mean 2	2SE2	Mean 3	2SE3
B	4.46	2.424	1.95	1.8	3.5	3
K	11.71	4.76	11.23	4.24	4.96	3.2
CL3	Mean 1	2SE1	Mean 2	2SE2	Mean 3	2SE3
B	6.92	3.152	1.86	1.92	4.13	2.96
K	16.93	5.24	12.7	5.36	6.46	3.8
CL4	Mean 1	2SE1	Mean 2	2SE2	Mean 3	2SE3
B	7.15	3.84	1.84	1.48	7.33	4.48
K	16.21	6.8	11.31	5.6	6.53	3.88
AM	Mean 1	2SE1	Mean 2	2SE2	Mean 3	2SE3
B	6.66	1.572	6.35	0.56	6.91	0.576
K	6.13	0.96	6.46	0.76	6.92	1.16

Table S2 Trait-specific means and one standard error for individual genotypes from each of the three experimental populations (Klamath_{SGT}, Klamath_{LGT}, and Lake Marie_{SGT}) for each of the three phenotypic assays.

		Klamath _{SGT}				
SM	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
B	1.69111	0.03818	2.002	0.04151	1.87945	0.05222
D	1.964	0.05138	2.25222	0.06084	2.12817	0.08709
F	1.90692	0.03181	2.26423	0.02587	2.16781	0.03986
G	2.16667	0.03306	2.39375	0.0302	2.21358	0.02937
H	2.04346	0.02675	2.26846	0.04668	2.23753	0.08957
I	1.838	0.05454	2.16115	0.04423	2.11942	0.07424
J	1.86609	0.03567	2.22	0.02806	2.0487	0.03303
K	1.8731	0.02682	2.25154	0.01834	2.14786	0.03614
M	1.90833	0.03241	2.18111	0.01957	2.01301	0.03589
CL1	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
B	2.3125	0.62464	3.4	0.61579	1.1	0.38406
D	2.71429	1.35742	5.16667	1.0918	5.11111	1.41421
F	3.30769	0.44732	5.03846	0.62865	4.25	0.67259
G	5.13333	0.7235	6.08333	0.88256	5	0.80204
H	2.09091	0.55824	3.92308	0.7679	2.77778	0.82776
I	3.25	0.88585	2.88462	0.66978	6.66667	1.10554
J	2.43478	0.70462	4.88889	0.7286	5.88889	0.60668
K	5.21429	0.677	6.19231	0.50277	4.72222	0.8965
M	1.95833	0.66667	2.96296	0.58465	3.04348	0.71371
CL2	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
B	3	1.12423	7.04	1.1878	1.25	0.44741
D			12.77778	2.0473	11.66667	2.85774
F	11.16	0.86453	10.65385	1.72118	6.66667	1.47263
G	13.93103	1.29137	17.25	1.38444	11.8	0.99594
H	9.89474	1.44338	8.26923	1.1912	6.88889	4.36102
I	7.6	1.29634	11.96154	1.69082	12.66667	2.35702
J	7.09091	1.626	11.25926	1.95059	12.11111	1.65412
K	10.18519	0.77885	13.57692	1.38183	6.83333	1.23228
M	6.45455	0.79722	14.22222	1.05902	7.30435	1.03884
CL3	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
B	4.27273	1.04062	7.16	1.71963	4.05	1.46527
D			19.22222	3.80614	14.875	3.53273
F	11.875	1.41966	10.84615	2.1916	3.41667	1.01673
G	17.2069	1.92058	20.875	1.88915	14.25	1.56072

H	10.88889	1.66188	14.03846	2.31449	4.66667	2.27303
I	6.77778	2.84392	15.76923	2.74586	12.16667	5.82142
J	8.21053	1.66607	19.92593	2.24906	14.77778	1.56125
K	11.42308	0.9857	16.80769	1.55982	6.72222	1.64227
M	10.47059	1.1589	15.92593	1.77049	8.30435	1.0526

CL4	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
B	2.90909	1.06919	11.24	1.73813	4.45	1.09525
D			20.11111	3.03243	17.375	4.39922
F	13.30435	1.58224	9.53846	2.39419	3.08333	1.08536
G	18.42857	1.85623	22.20833	2.7821	14.65	1.87697
H	11.27778	1.44161	16.30769	2.09404	10.11111	5.69763
I	9.625	3.47906	15.92308	3.68236	14	5.60753
J	12.57895	1.63849	23.03704	2.74621	17.88889	1.81621
K	12.95833	0.80592	20.03846	1.83246	12.26667	2.32578
M	12.41176	1.13025	15.96296	2.00385	11.95238	1.49317

AM	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
B	7.72305	0.68432	6.0468	0.27039	6.58142	0.38831
D	7.508	0.85732	6.52889	0.39209	7.36205	0.74563
F	5.94724	0.2059	5.95731	0.23634	6.47563	0.17033
G	6.38018	0.25681	6.23667	0.19481	6.72361	0.26628
H	6.52181	0.39364	6.62192	0.28672	7.4095	1.16907
I	6.86785	0.42673	5.97577	0.24111	6.83149	0.51572
J	6.55759	0.34808	6.68259	0.56116	6.93879	0.33322
K	6.53339	0.14558	6.12115	0.16449	6.83465	0.39046
M	6.75191	0.4088	5.89111	0.19927	6.46791	0.24094

Klamath_{LGT}

SM	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
D	1.98714	0.03649	2.29962	0.02321	2.14567	0.05293
F	1.96633	0.02622	2.22759	0.03078	2.04506	0.03888
G	2.05852	0.03268	2.40533	0.02522	2.14567	0.04566
H	2.01889	0.03452	2.30095	0.02857	2.14567	0.07557
J	1.84786	0.03072	2.23067	0.02247	2.14661	0.03761
M	1.84174	0.03283	2.18214	0.0244	2.05927	0.05814

CL1	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
D	2	0.78707	4.23077	0.72815	4.54545	0.84092
F	3.03333	0.49814	4.41379	0.50161	2.63158	0.53593
G	4.2963	0.71308	7.66667	1.51897	4.31579	0.64877

H	2.37037	0.43243	3.90909	0.61464	2.23077	0.61592
J	3.21429	0.84537	7.43333	0.49566	7	1.11665
M	2.90476	0.7962	4.03571	0.63036	1.66667	0.97068
CL2	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
D	6.5	1.82312	14.42308	2.04383	10.36364	2.2753
F	11.34483	1.10771	12.17241	1.23149	5.31579	1.0341
G	12.22222	1.28425	18.33333	1.58785	11.36842	1.10417
H	8.125	0.90669	6.54545	1.33452	4	1.50321
J	7.39286	1.14677	18.33333	1.16063	14	1.87248
M	7.85714	1.07067	13.71429	0.78201	8.05882	0.85643
CL3	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
D	10.22222	1.94414	19.03846	2.08811	11.27273	2.92499
F	13.2963	0.84209	10.13793	3.07336	4.68421	0.90418
G	16.22222	1.51891	21.36667	1.5178	12.33333	1.38919
H	11.58333	0.95769	9.95455	2.13591	4.38462	1.17829
J	8.96296	1.74081	20.86667	1.22189	17.35	1.98078
M	10.31579	0.61281	16.14286	1.52694	11.82353	1.26227
CL4	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
D	8.28571	5.62028	22.80769	2.63607	13.6	3.78871
F	14.66667	1.86017	13.44828	3.36754	4.55556	1.8407
G	19.14815	1.26808	22.93333	2.41668	12.66667	1.83317
H	10.80952	1.79025	14.27273	3.74578	3.84615	2.03369
J	13.51852	1.67346	28.73333	2.64738	19.85	2.21464
M	8.5	1.49642	16.92857	1.99864	12.52941	3.21636
AM	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
D	7.54688	0.61291	6.17923	0.17816	6.65861	0.39264
F	6.52135	0.29006	5.87034	0.21236	6.82576	0.52245
G	6.27675	0.2578	5.97133	0.16632	6.78611	0.45291
H	6.46136	0.31493	6.38091	0.23506	7.01283	0.64546
J	6.43364	0.30622	6.07633	0.1188	6.74281	0.23265
M	6.643	0.26849	5.99571	0.12323	6.98517	0.28321

Marie

SM	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
B	1.75833	0.03236	1.885	0.0206		
F	1.82481	0.02514	1.90091	0.05339	1.87289	0.02986
H	1.75933	0.03612	1.83353	0.03047		
I	1.82714	0.04104	1.80889	0.0358		

K	1.84704	0.02724	1.99037	0.02055	1.92828	0.0273
L	1.98522	0.02253	2.04842	0.0637		
M	1.82875	0.03742	1.8725	0.02862		
CL1	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
B	2.36842	1.00236	0.35714	0.33369		
F	5.51852	0.45134	4.14286	1.72773	2.07143	0.67234
H	2.46667	0.5529	1.11765	0.52723		
I	3.6	1.09929	0.6	0.48553		
K	4.37037	0.3849	4.7037	0.4598	3.43478	0.58994
L	5.08333	0.81022	2.89474	1.33074		
M	3.125	0.83553	0.92308	0.39099		
CL2	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
B	5.52632	0.99673	0.57143	0.22527		
F	13.22222	1.21144	6.57143	3.20123	5.92857	1.39654
H	3.6	0.89785	1.35294	0.42462		
I	7.93333	1.99712	2.2	1.2333		
K	11.96296	1.29159	12.07407	1.33824	8.54545	1.27277
L	11.625	1.47148	7.89474	1.49436		
M	6.25	0.85928	1.30769	0.81009		
CL3	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
B	5.42105	1.10972	1.42857	0.69607		
F	12.59259	1.55431	3.85714	2.01914	7.28571	1.01835
H	4.06667	0.88481	1.35294	0.45659		
I	8.86667	2.73351	3.1	1.25681		
K	11.77778	1.30037	14.25926	1.66539	8.18182	1.63249
L	10.41667	1.92982	10.15789	3.62889		
M	8.5	1.34725	1.38462	0.64659		
CL4	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
B	4.78947	1.20652	1.28571	0.62703		
F	14.62963	1.56889	5.35714	2.07485	5.35714	1.46986
H	3.6	1.12838	0.70588	0.38315		
I	7.26667	2.62354	3.5	1.88542		
K	15.44444	1.81525	17.33333	1.80662	8.5	1.85029
L	9.58333	1.82357	7.31579	2.89184		
M	7.625	2.18111	1.84615	1.27127		
AM	Mean 1	SE1	Mean 2	SE2	Mean 3	SE3
B	6.845	0.38082	6.55	0.79418		
F	6.48538	0.23584	6.47	0.38744	6.71091	0.39535

H	6.78667	0.84759	6.64765	0.52907		
I	6.78286	0.35139	6.54111	0.33587		
K	5.52148	0.91218	6.31259	0.14336	6.55009	0.11752
L	6.36682	0.20068	6.56737	0.34794		
M	6.69	0.29772	6.6325	0.56854		

Table S3 The number of generations of divergence (GOD), trait-specific genetic variances (VG) and one standard error of the genetic variance for individual genotypes from each of the three experimental populations (Klamath_{SGT}, Klamath_{LGT}, and Lake Marie_{SGT}) for each of the three phenotypic assays.

Klamath _{SGT}									
SM	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
B	11	0	0.00463	24	0.01072	0.00769	58	0.01061	0.00969
D	12	0.0036	0.0087	27	0.01694	0.01313	62	0.0057	0.01613
F	14	0.00032	0.00485	31	0	0.00294	68	0.004	0.00645
G	11	0.0062	0.00478	28	0	0.00337	63	0.00013	0.00358
H	13	0	0.00362	28	0.01441	0.00881	61	0	0.01147
I	13	0.00865	0.01144	28	0.00969	0.0081	60	0.00284	0.00802
J	14	0	0.00529	30	0.00319	0.00332	67	0.00543	0.0045
K	13	0.00055	0.00361	29	0	0.00124	64	0	0.00454
M	12	0.00205	0.0048	29	0.00094	0.00169	65	0	0.00517
CL1	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
B	11	0.67027	1.56848	24	0	1.74764	58	0.21922	0.57401
D	12	1.05556	5.80913	27	3.63636	4.40911	62	0	3.64095
F	14	0	0.80324	31	0.88528	1.92736	68	0.20833	1.98974
G	11	0	2.6108	28	3.7996	2.97242	63	1.80837	2.37236
H	13	0.61744	1.59833	28	0	1.66246	61	0	0.92617
I	13	2.64833	3.06826	28	1.10444	1.98736	60	0	0.91961
J	14	1.58949	2.18545	30	0	2.27256	67	0.61111	1.66505
K	13	1.3608	2.15364	29	0	1.05822	64	2.2037	2.64732
M	12	0	1.65521	29	1.04321	1.47896	65	2.28013	1.96756

CL2	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
B	11	4.71748	4.73966	24	0	6.23078	58	0	0.74921
D	12			27	7.09519	16.45037	62	11.66667	16.55406
F	14	0	2.81939	31	8.24783	14.18914	68	0	7.55089
G	11	0	5.92863	28	0	8.4872	63	0	3.85083
H	13	11.98125	9.60638	28	0	6.03001	61	31.44444	38.227
I	13	5.42988	5.79037	28	7.79791	12.55674	60	1.66667	8.32407
J	14	12.91625	11.12595	30	7.87037	16.97681	67	18.79012	10.99308
K	13	3.14634	2.70407	29	6.26562	8.24597	64	0	4.14217
M	12	0.47635	3.11998	29	3.87346	4.79727	65	0	3.30408

CL3	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
B	11	1.90012	3.39643	24	0	11.32138	58	8.11374	7.68881
D	12			27	76.2365	51.00383	62	17.96349	25.47911
F	14	3.47412	9.29585	31	25.08792	21.57292	68	0	4.03328
G	11	17.61942	16.46678	28	1.64881	15.69624	63	4.74097	9.28993
H	13	17.00918	12.75911	28	5.00716	25.05459	61	6.88889	10.52295
I	13	26.57576	28.44506	28	50.31203	30.47006	60	0	24.04343
J	14	7.68766	12.92731	30	14.49074	22.00781	67	10.96914	10.15621
K	13	0	3.96699	29	0	8.76744	64	7.05926	8.92313
M	12	0	4.85086	29	8.41049	13.71423	65	0	3.22635

CL4	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
B	11	2.55733	3.4845	24	0	14.88789	58	0	4.83578
D	12			27	0	30.05736	62	12.75079	42.08296

F	14	0	8.62149	31	23.79222	26.38406	68	5.85714	4.48788
G	11	12.65199	15.8692	28	0	19.77745	63	0	9.10526
H	13	0	12.61747	28	6.12314	20.15946	61	29.96296	67.99151
I	13	55.71154	40.07099	28	90.52304	54.79745	60	0	27.04648
J	14	0	12.83642	30	0	24.25649	67	10.0679	14.27202
K	13	0	3.4603	29	0	13.54066	64	12.04771	18.65331
M	12	2.77539	5.61912	29	5.8642	18.29196	65	0	6.52289

AM	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
B	11	0	1.45661	24	0	0.34591	58	0	0.44262
D	12	0	2.24522	27	0.04247	0.65146	62	0	0.69429
F	14	0.03682	0.19879	31	0	0.22505	68	0	0.11508
G	11	0	0.25139	28	0.15741	0.14706	63	0.02329	0.29191
H	13	0.52142	0.72764	28	0	0.31844	61	2.64759	2.73388
I	13	0.67891	0.68003	28	0.04236	0.27405	60	0.23072	0.37697
J	14	0.43806	0.5344	30	0	1.34215	67	0	0.36319
K	13	0	0.10559	29	0	0.12777	64	0.59937	0.48668
M	12	0.37698	0.75546	29	0.14571	0.16886	65	0	0.2077

Klamath_{LGT}

SM	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
D	7	0.00215	0.00637	17	0.00106	0.00243	27	0	0.00641
F	8	0.00026	0.00347	14	0.00604	0.00411	21	0	0.00464
G	7	0.00397	0.00454	13	0.00345	0.00279	20	0.00634	0.00763
H	8	0.00203	0.00587	14	0	0.00253	22	0.01461	0.01676
J	10	0	0.00412	13	0	0.00202	21	0.00034	0.00579

M	9	0	0.00461	17	0	0.00214	29	0.01186	0.01089
CL1	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
D	7	0	2.85228	17	0	1.79968	27	0	1.66733
F	8	0.05556	1.25804	14	0.44751	1.21967	21	0.46078	1.12272
G	7	1.62346	2.19109	13	18.79877	9.80865	20	0.51542	1.67377
H	8	0	0.78566	14	0.35714	1.65759	22	0.51493	1.18589
J	10	2.87216	3.26131	13	0	1.18363	21	3.73545	4.54557
M	9	2.59215	2.49171	17	0	1.38728	29	2.45185	3.11906
CL2	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
D	7	0	17.23978	17	21.77046	17.21842	27	0	10.16958
F	8	3.58122	5.74376	14	0	7.41623	21	0	3.67561
G	7	4.60185	7.19215	13	10.19136	11.4038	20	0	5.13406
H	8	1.57059	4.13008	14	0	6.1567	22	0	5.01108
J	10	0	6.35242	13	0.3679	6.81771	21	11.24339	12.69729
M	9	0	4.67332	17	0.66477	3.05535	29	1.05404	2.57458
CL3	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
D	7	13.46875	13.24163	17	11.51225	19.20428	27	0	16.0853
F	8	0	3.37041	14	78.18973	40.30733	21	0	3.27047
G	7	0	9.25692	13	0	9.84922	20	0	5.94668
H	8	0	4.19712	14	0	19.56205	22	0.41978	4.69168
J	10	9.78126	14.20807	13	3.25926	7.10506	21	15.55173	13.9879
M	9	0	1.24262	17	5.63494	11.14281	29	3.3303	5.41893
CL4	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
D	7	0	225.2659	17	4.62771	32.83617	27	0	36.75779

F	8	21.22666	15.15817	14	79.30437	48.82245	21	0	12.04766
G	7	0	7.15185	13	16.31852	27.30084	20	8.82115	12.63877
H	8	10.80329	12.87009	14	54.44286	55.50351	22	8.14366	12.46742
J	10	0	14.30439	13	0	20.42342	21	4.18271	19.66714
M	9	0	9.87739	17	0	15.76466	29	43.16061	33.09334

AM	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
D	7	1.78587	1.60833	17	0	0.13735	27	0.31066	0.38791
F	8	0.11333	0.41094	14	0.2558	0.1978	21	0	0.98734
G	7	0	0.26656	13	0	0.12841	20	0.81169	0.73144
H	8	0.117	0.49791	14	0	0.16037	22	1.55539	1.19434
J	10	0	0.34897	13	0	0.04468	21	0.18682	0.19556
M	9	0	0.3254	17	0	0.07071	29	0	0.23553

Marie

SM	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
B	12	0.01423	0.01715	27	0.04173	0.06589			
F	14	0.00068	0.00292	32	0.01032	0.00842	57	0.00145	0.00272
H	14	0.00389	0.00497	30	0.00349	0.00299			
I	12	0.00241	0.00715	28	0	0.00319			
J	18	0.00298	0.00313	36	0	0.00149			
K	13	0	0.00191	29	0.02116	0.01448	66	0.00246	0.00327
M	12	0.00221	0.00493	27	0	0.00452			
CL1	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
B	12	9.2037	8.67665	27	6.71739	6.431			
F	14	0	0.69411	32	9.20133	8.585	57	0	0.87258

H	14	0.73882	1.19822	30	0	0.63302			
I	12	0	3.96674	28	0.08784	0.6793			
J	18	0	0.64209	36	0	1.01125			
K	13	2.48413	2.56716	29	7.56209	6.36515	66	1.01496	1.5472
M	12	0	1.7041	27	1.0473	0.64683			
CL2	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
B	12	19.93519	17.86768	27	40.47101	37.59285			
F	14	0	4.99088	32	27.80247	29.76014	57	1.41952	6.28477
H	14	0.90921	3.4171	30	0	0.46764			
I	12	1.49069	17.80141	28	2.00676	4.01669			
J	18	0	5.84921	36	4.14198	7.92738			
K	13	2.5873	9.24041	29	9.85948	8.01073	66	1.21352	7.9515
M	12	0	1.94786	27	1.91216	3.54373			
CL3	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
B	12	0	14.54704	27	3.65217	11.19872			
F	14	8.98148	10.26047	32	8.41026	12.14323	57	0	3.34979
H	14	0	2.97304	30	0.42525	0.71062			
I	12	29.64295	27.73263	28	2.0473	4.17883			
J	18	2.97222	7.62431	36	13.17593	11.49367			
K	13	0	10.5852	29	70.7549	46.98801	66	10.4229	11.5278
M	12	0	7.00063	27	1.06081	2.33114			
CL4	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
B	12	2.10185	15.29273	27	131.3623	118.5344			
F	14	0	11.12182	32	7.83357	12.9765	57	0	5.78328
H	14	4.98487	4.67624	30	0.43889	0.48224			

I	12	26.85638	25.58624	28	0	6.34153			
J	18	5.47222	14.90671	36	2.19753	15.29665			
K	13	0	9.90451	29	22.73203	31.25187	66	0.73347	17.22923
M	12	0	11.47289	27	12.13851	6.78263			
AM	GOD 1	VG 1	SE1	GOD 2	VG 2	SE2	GOD 3	VG 3	SE3
B	12	1.05695	1.38772	27	2.18494	3.23428			
F	14	0.04329	0.2617	32	0.2201	0.48058	57	0.23385	0.47953
H	14	2.91159	2.62771	30	0.17802	1.02716			
I	12	0	0.45979	28	0.2054	0.28981			
J	18	1.94543	3.68016	36	0	0.07784			
K	13	0	0.12012	29	0.2934	0.45727	66	0	0.04555
M	12	0	0.29046	27	0.48353	1.74699			

Table S4 Trait-specific estimates of ΔV (VAR) and ΔM (MEAN) for individual genotypes from each of the three experimental populations (Klamath_{SGT}, Klamath_{LGT}, and Lake Marie_{SGT}).

Population	Genotype	SM _{VAR}	SM _{MEAN}	CL1 _{VAR}	CL1 _{MEAN}	CL2 _{VAR}	CL2 _{MEAN}	CL3 _{VAR}	CL3 _{MEAN}	CL4 _{VAR}	CL4 _{MEAN}	AM _{VAR}	AM _{MEAN}
Marie	B	0.00000	0.00870	0.00790	-0.13400	0.00290	-0.33100	0.05230	-0.26600	0.02640	-0.23300	0.01140	-0.02000
	F	0.00005	0.00110	0.00021	-0.08020	0.00003	-0.16700	0.00010	-0.09690	0.04900	-0.20250	0.00790	0.00500
	H	0.00014	0.00440	0.00540	-0.08400	0.00190	-0.14100	0.01350	-0.17000	0.02210	-0.18100	0.02180	-0.00800
	I	0.00002	-0.00130	0.00304	-0.18800	0.07400	-0.35800	0.13800	-0.36100	0.09700	-0.23600	0.00658	-0.01500
	K	0.00003	0.00120	0.00940	-0.01990	0.03760	-0.07580	0.20250	-0.08170	0.04060	-0.15950	0.00009	0.01290
	L	0.00030	0.00380	0.23700	-0.13700	0.31900	-0.23400	1.29000	-0.01630	0.47800	-0.14100	0.00570	0.01250
	M	0.00003	0.00270	0.03600	-0.14700	0.05210	-0.32900	0.03700	-0.47500	0.40300	-0.38500	0.00820	-0.00400
Klamath _{SGT}	B	0.00008	0.00300	0.00400	-0.03600	0.00200	-0.07300	0.12800	-0.01400	0.01050	-0.00037	0.00000	-0.00700
	D	0.00020	0.00300	0.01900	0.03800	0.20000	-0.03200	0.50900	-0.12400	0.16300	-0.07700	0.00026	0.00500
	F	0.00004	0.00400	0.00700	0.01500	0.02400	-0.08500	0.03400	-0.16300	0.10700	-0.18700	0.00006	0.01060
	G	0.00001	-0.00010	0.04200	-0.00600	0.00000	-0.06100	0.09700	-0.08100	0.01800	-0.08800	0.00180	0.00780
	H	0.00010	0.00500	0.00100	0.01100	0.27100	-0.06500	0.50200	-0.00400	0.04780	-0.10900	0.01690	0.01790
	I	0.00012	0.00500	0.00600	0.07900	0.07900	0.10600	0.32100	0.11200	0.42300	0.08300	0.00430	0.00400
	J	0.00008	0.00100	0.01100	0.05800	0.30200	0.08400	0.20200	0.08400	0.12900	0.07300	0.00086	0.00720
	K		0.00400	0.02500	-0.01400	0.03200	-0.07300	0.08500	-0.10200	0.12500	-0.00300	0.00470	0.00510
M	0.00002	0.00200	0.03400	0.01800	0.01700	0.00300	0.01270	-0.06300	0.02120	-0.01600	0.00120	0.00180	
Klamath _{LGT}	D	0.00004	0.00900	0.00000	0.12900	0.23500	0.21500	0.27000	0.11400	0.07800	0.15700	0.00700	-0.03200
	F	0.00014	0.00700	0.01700	0.18200	0.03170	-0.48200	0.17100	-0.66400	0.75700	-0.78700	0.01300	0.01100
	G	0.00030	0.01030	0.13000	-0.01200	0.16200	-0.10700		-0.34100	0.48600	-0.47200	0.01100	0.03100
	H	0.00020	0.01500	0.02100	-0.00700	0.02100	-0.29800	0.01500	-0.51600	0.73600	-0.51200	0.01700	0.03500
	J		0.02000	0.07000	0.40700	0.25600	0.45900	0.58100	0.48500	0.11800	0.37300	0.00320	0.05000
	M	0.00010	0.01300	0.06300	-0.07000	0.03500	-0.05600	0.10700	0.09300	0.72800	0.24500		0.02500