Adaptive Plasticity in Female Mate Choice Dampens Sexual Selection on Male Ornaments in the Lark Bunting

Alexis S. Chaine, *et al.*

Science **319**, 459 (2008);
DOI: 10.1126/science.1149167

Adaptive Plasticity in Female Mate Choice Dampens Sexual Selection on Male Ornaments in the Lark Bunting

Alexis S. Chaine[1,2]* and Bruce E. Lyon[1]

Theory on the evolution of ornamental male traits by sexual selection assumes consistency in selection over time. Temporal variation in female choice could dampen sexual selection, but scant information exists on the degree to which individual female preferences are flexible. Here we show that in lark buntings sexual selection on male traits varied dramatically across years and, in some cases, exhibited reversals in the direction of selection for a single trait. We show that these shifts are probably because of flexibility in mate choice by individual females and that they parallel shifts in the male traits that predict female reproductive success in a given year. Plasticity in choice and concomitant reversals in mating patterns across time may weaken the strength of sexual selection and could maintain genetic variation underlying multiple sexual ornaments.

Sexual selection for exaggerated secondary sexual traits arises from two different mechanisms that result in correlations between male traits and mating success—male-male competition for access to females and female mate choice (*1*, *2*). These mechanisms can be difficult to disentangle (*3*), but direct female choice for extravagant male traits has been shown in many taxa (*2*). Ornament evolution via female choice requires that females prefer an extreme expression of a male trait, that trait preferences are concordant among females in a population, and that these preferences are fairly consistent across time (*4*, *5*). Changes in the ecological or social environment could favor flexibility in female preferences (*6*, *7*), but information about the temporal consistency of female choice is currently lacking (*6*, *8*). Plasticity in female preferences could have major effects on the strength and outcome of sexual selection (*9*) and, potentially, could slow trait exaggeration (*10*).

We investigated the dynamics of pairing patterns and mate choice in the lark bunting, *Calamospiza melanocorys,* a migratory songbird breeding on the short-grass prairie of Colorado. Sexual selection is potentially strong in lark buntings, because many males fail to attract a social mate [~45% of territorial males (*11*)] because of a male-biased breeding sex ratio coupled with social monogamy and because extra-pair paternity is common [25% of young and 47% of broods (*11*)], but variable among males. To assess the dynamics of sexual selection, we studied five independent male plumage traits and three measures of size (Fig. 1)—body color, proportion of black versus brown feathers separately on the rump and the rest of the body, wing patch size, wing patch color, body size, beak size, and residual mass (*12*). To examine sexual selection on these male traits, we assessed both the social pairing success of color-banded males in each breeding season, as well as their total annual fitness, using microsatellite parentage analysis (*12*).

In territorial birds, it can be difficult to distinguish between direct female choice for male traits and female choice for territory features correlated with male traits (i.e. male dominance badges). Male lark buntings, however, are only weakly territorial until mate acquisition, at which time the territory is no longer defended nor respected by other males [new males begin displaying on the former territory (*11*)]. Display territories are not used for feeding by either males or females (including offspring feeding), but females nest near or on the display territory, so we quantified territory quality as the density of woody shrubs available for use as nest cover (*12*).

In each of the 5 years of this study, plumage or size characteristics of males were associated with total male fitness [number of within- and extra-pair fledglings sired (*12*) (Fig. 2)], which indicated significant potential for sexual selection on those male traits. However, a significant effect of year on the traits that correlate with male fitness indicated that the specific traits under sexual selection varied among years [according to a generalized linear model (GLM): full model $F_{3,380} = 5.19$, $P = 0.001$; effect of year: $F_{4,379} = 7.52$, $P < 0.001$ (*12*)]. Examination of these patterns revealed dramatic changes across years in the suites of traits that predicted male fitness, and moreover, no two years showed similar patterns of male traits associated with fitness (Fig. 2).

We observed two distinct types of change in the pattern of selection on male traits across years. First, some male traits were under strong selection in some years, but showed very weak selection or no selection in other years [beak size and rank body color (Fig. 2)]. Cubic splines illustrate the specific form and intensity of phenotypic selection on male traits each year (*12*, *13*) and demonstrate the occurrence of this on-off pattern of selection

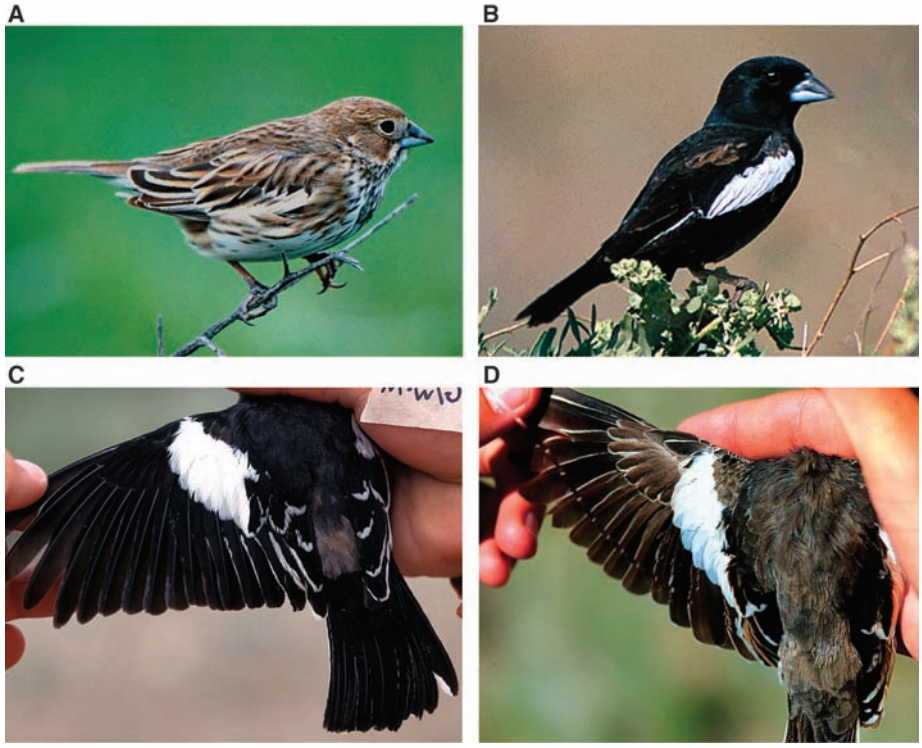

Fig. 1. Plumage traits of lark buntings. (**A**) Females are brown with dull white wing patches, whereas (**B**) males are generally black, but often have patches of brown feathers that vary in size and distribution (note patch above wing). The conspicuous white wing patches vary in both size and color among males and are both larger and brighter than those of females. There is considerable variation in color between (**C**) black males and (**D**) gray ones. Likewise, the proportion of dark versus brown feathers varies among males on both the body [(C) versus (D)] and rump (C) relative to other body parts.

[1]Department of Ecology and Evolutionary Biology, University of California, Santa Cruz, CA 95064, USA. [2]Laboratoire d'Evolution et Diversité Biologique, Station d'Ecologie Expérimentale du CNRS à Moulis, USR 2936, 09200 Saint-Girons, France.

*To whom correspondence should be addressed. E-mail: alexis.chaine@ecoex-moulis.cnrs.fr

for several traits [colored lines (Fig. 3)]. Similar effects have been shown in other species (*14*, *15*). Second, three traits—body size and the percentage of black feathers on both the rump and the rest of the body—showed a positive association with male fitness in 1 year and a negative association in another year. These reversals from positive to negative selection gradients were significant (GLM: year × body size $F_{4,379} = 3.56$, $P = 0.007$; year × rump% $F_{4,379} = 3.25$, $P = 0.012$; year × body% $F_{4,379} = 3.08$, $P = 0.016$) and indicate that dramatic shifts in selection occur across years [(Fig. 2) and colored lines in (Fig. 3)]. The reversals in selection on male traits we document here provide a sexual selection parallel to oscillating natural selection described for Darwin's finches (*16*).

To understand the underlying cause of variable selection on male traits, we investigated a key component of male fitness: acquisition of a social mate. The general pattern of selection on male traits through mate acquisition (Fig. 4) was similar to overall selection on male traits (Fig. 2)—there was a significant association between male traits and mate acquisition, but the traits of successful males varied across years [GLM: full model $F_{3,380} = 10.22$, $P < 0.001$; effect of year: $F_{4,377} = 4.13$, $P = 0.002$; (*12*)]. Some traits were under selection because of mate acquisition in some years but not others (percentage black feathers on the body, beak size, and residual mass). Two traits—wing patch size and rank body color—showed changes in the direction of selection across years (GLM: year × wing patch size $F_{4,415} = 3.24$, $P = 0.012$; year × rank color $F_{4,415} = 3.29$, $P = 0.011$). A randomization test (*12*) indicated a close correspondence between the traits under selection because of total fitness and those under selection because of mate acquisition [Fig. 2 versus 4, $P < 0.001$, (*12*)]. The striking similarity in patterns of selection on male traits through total male fitness and through mate acquisition occurs because the majority of offspring are sired within the social pair [75% (*11*)] and suggests that across-year variation in social mating success is a major driver of variable selection on male traits.

Several observations suggest that temporal fluctuations in social mating patterns are more likely to be because of changes in female choice for male traits than changes in traits that influence competition among males for high-quality territories that females might choose. First, territory quality (*12*) was never associated with mate acquisition [i.e., was not selected by Akaike's Information Criterion (AIC) models for all years combined or for any individual year; partial $P > 0.2$ in all cases] and was therefore removed from all selection models. Second, we determined whether traits previously found to be important to male-male competition (*11*) differed across years in their importance as dominance signals. We did this using both observational and experimental methods, but in no case did the male traits associated with social dominance change across years in a manner that could explain the dynamic patterns of mate acquisition we describe here

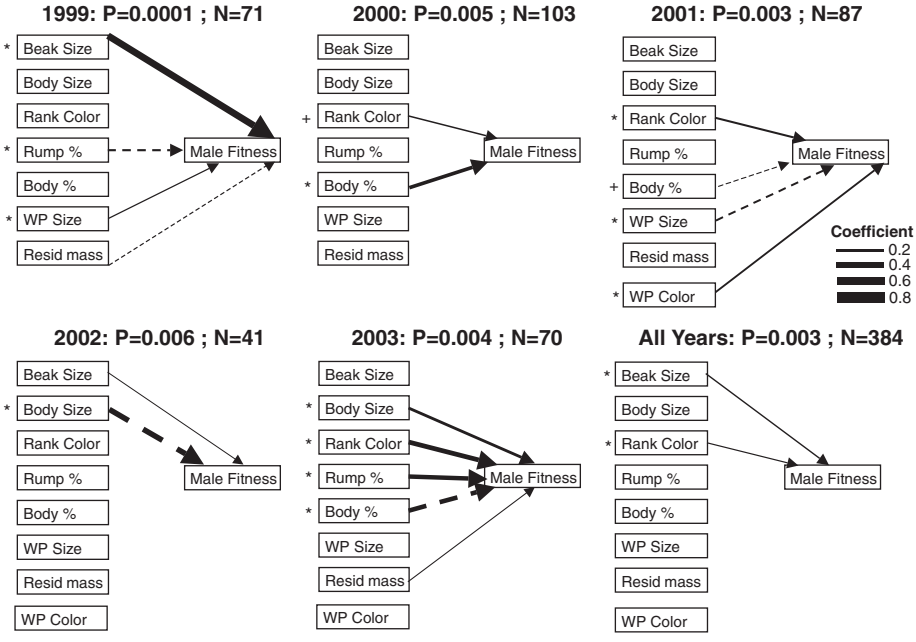

Fig. 2. Male traits associated with total male fitness (number of sired within and extra-pair chicks that fledged) show dramatic across-year variation. Year, *P* value, and sample size from the final GLM (*12*) are above each diagram. Thickness of arrows between traits and fitness indicate the partial correlation coefficient (selection gradient). Dashed and solid arrows are negative and positive relationships, respectively. Traits include three morphometric measures (beak size, body size, and residual mass) and five plumage measures: body feather color (rank color), percentage black feathers on the rump (rump%) and the rest of the body (body%), wing patch size (WP size), and wing patch color (WP color). Partial effect in GLMs designated as $*P < 0.05$, $+P < 0.1$.

(*12*). Furthermore, the level of male aggression in response to a standardized stimulus did not differ across years, which suggested that the intensity of male-male competition did not vary across years (*12*).

Two lines of evidence indicated that population-level changes in female mate choice across years were because of plasticity in individual female preferences across years rather than age-related or other demographic shifts (*12*). Females that were observed in 2 or more years and that did not stay with the same social mate (*12*) were used to estimate the degree of individual consistency [repeatability, r_t (*17*)] in mate choice (19 females whose mates were also caught: 17 in 2, 1 in 3, and 1 in 4 years). Mate choice (traits of her chosen mates) was not significantly repeatable across time (repeatability: all traits df = 2.2,19, $r_t < 0.18$; $P > 0.2$), which suggests substantial plasticity of female choice. Indeed, most females showed considerable change in the traits of their mates across years (fig. S1). Changes in the mate choice decisions of individual females will only sum to population-level dynamic sexual selection if most females show similar patterns of plasticity and response, a pattern that implicates the influence of external factors such as the social or ecological environment (*18*, *19*).

We investigated the possibility that plasticity in choice is adaptive because it allows a female to choose a male or territory character that will maximize her fitness in each year. This hypothesis predicts that females target male or territory characters that predict fitness benefits to them in a given year, that different traits serve as fitness indicators in different years, and that changes in female preference across years correspond nonrandomly with the changes in fitness-indicator traits. In most years, one or more male traits were correlated with nesting success (a measure of female fitness, fig. S2) making them potential indicator traits, and different traits were predictors of nesting success in different years (fig. S2). Randomization tests determined that there were significantly more matches than expected by chance between the traits of males chosen by females and the male traits associated with nesting success within years [four of seven male traits correlated with nesting success were also preferred by females, $n = 76$ total trait-year possibilities, $P = 0.005$; see (*12*)]. This result suggests that flexible female choice enables females to track temporal variation in the traits that predict enhanced nesting success. However, it remains unknown whether the specific traits we measured or correlates of those traits are the actual targets of female choice (*20*).

Sexual selection by female choice requires that the cumulative effects of female preferences be fairly consistent over time (*1*, *2*), and models of sexual selection with consistent choice predict extreme exaggeration of male traits (*4*, *5*). However, if female choice varies across years, phenotypic selection for male trait exaggeration could be dramatically reduced or even eliminated, as has been suggested for temporal variation in natural selection (*21*, *22*). We investigated the

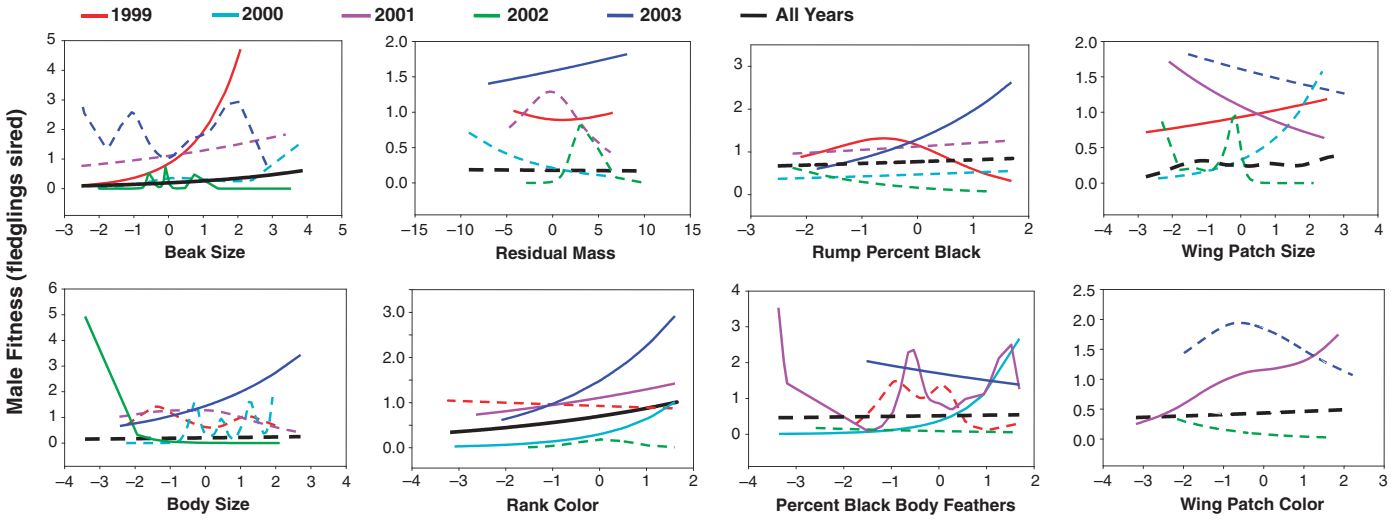

Fig. 3. Cubic splines reveal the dynamic nature of selection on male traits because of male fitness (number of fledglings sired within pair and extra-pair) across years. Colored splines represent selection on male traits in a given year; black splines represent selection in all years combined. Wing patch size and the percentage of black feathers on both the rump and rest of the body show marked reversals in the direction of selection, whereas rank body color and beak size show consistent positive relationships in all years. Solid lines depict traits that entered into selection models [models and *P* values in (Fig. 2); see (*12*)] for that year (or all years), whereas dashed lines were not significant. Only rank color and beak size show significant selection in the analysis using data from all years combined.

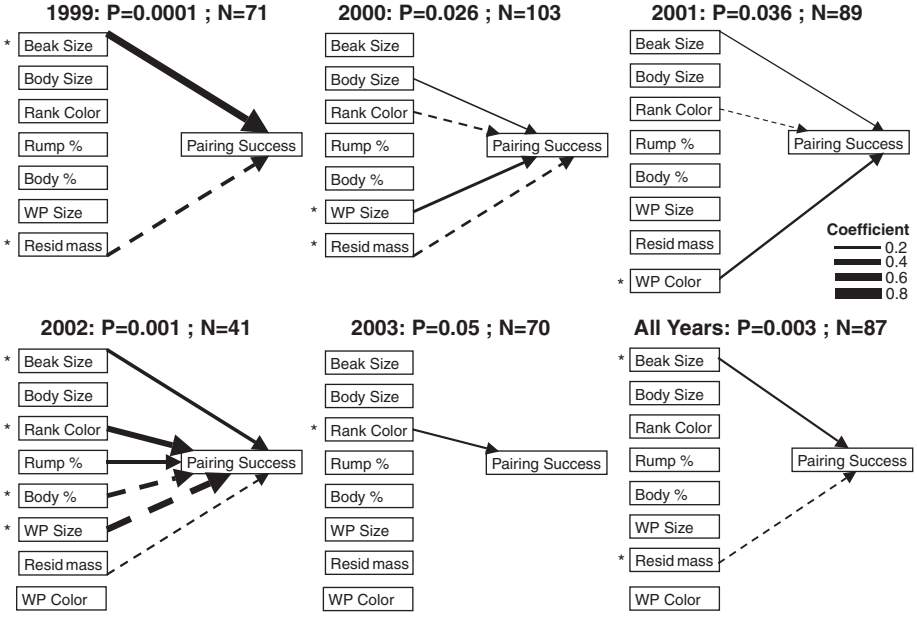

Fig. 4. Male traits associated with pairing success (mated or not mated) differ dramatically across years, presented as in (Fig. 2). Traits include three morphometric measures (beak size, body size, and residual mass) and five plumage measures: body feather color (rank color), percent black feathers on the rump (rump%) and rest of the body (body%), wing patch size (WP size), and wing patch color (WP color). Partial effect in GLMs designated as $*P < 0.05$.

impact of temporal scale on phenotypic selection by comparing analyses from individual years to an analysis of data combined across years [e.g. (Fig. 2)]. For most traits, phenotypic selection of all years combined was weak and only two traits—beak size and rank body color—were under significant selection as determined by overall male fitness patterns (Fig. 2). A similar pattern can be seen in selection on male traits through female choice of a social mate (Fig. 4). The striking effect that temporal scale has on the intensity and pattern of sexual selection can be seen by comparing selection splines on individual male traits from each year separately with those from all years pooled [colored versus black lines (Fig. 3 and fig. S3)]. By examining years separately, a very different picture of selection on male traits and female mate choice emerges from the standard approach obtained by summing across years. In lark buntings, selection on male traits is often strong within years, dynamic across years, but weak or nonexistent over a 5-year period.

Which time scale is the most appropriate for understanding sexual selection? We suggest that a longer time frame is most appropriate for predicting male trait evolution because annual male fitness measures used to estimate short-term sexual selection are unreliable when selection fluctuates across years. Short-term studies may thus prove insufficient for correctly understanding the strength and direction of sexual selection in species with variable sexual selection and flexible mate choice. In contrast, a short time frame is essential for understanding selection on female mating preferences. Only by examining female choice patterns within years was it possible to discover flexibility in choice and show the adaptive benefits of flexible choice to females.

Plasticity in female preferences has several implications for the process of sexual selection, the evolutionary dynamics of exaggerated traits, and the evolution of female choice itself. As a parallel to natural selection under variable environmental conditions, flexible female choice and dynamic sexual selection could make assessment of selection and predictions for male trait evolution unreliable (*22*) and may also provide a mechanism for the preservation of genetic variation in traits (*23*). In addition, most models of sexual selection assume that the evolution of exaggerated traits is stabilized by the costs that exaggerated traits incur (*5*, *24*). In taxa with flexible female preferences, however, choice itself may result in stabilizing selection on exaggerated traits. Finally, flexible female choice may provide an explanation for the evolution and maintenance of multiple male ornaments. Flexible choice should allow a female either to choose a mate that best complements her needs in a given year, where such needs change, or to track the best signals

when signal content changes over years because of changes in the physical or social environment. When females vary their preferences across years, the expression of more than one quality by a male would give him a mating advantage across breeding seasons by providing broad appeal under unpredictable breeding conditions. Under this scenario, selection should favor the evolution of new indicator traits that prove useful to females in some years and maintain existing multiple signals despite countervailing costs. Variation in female preferences may provide explanations for what is currently considered noise [e.g., unexplained variance, (*8*); low repeatability, (*25*, *26*)] in female choice. Testing for and incorporating temporal flexibility in female choice has the potential to greatly alter our perspectives on the process of sexual selection and trait exaggeration.

References and Notes

1. C. Darwin, *The Descent of Man and Selection in Relation to Sex* (Murray, London, UK, 1871).
2. M. Andersson, *Sexual Selection* (Princeton University Press, Princeton, NJ, 1994).
3. D. Westneat, I. Stewart, *Annu. Rev. Ecol. Evol. Syst.* **34**, 365 (2003).
4. P. O'Donald, in *Mate Choice* P. Bateson, Ed. (Cambridge University Press, Cambridge, UK, 1983), pp. 53–66.
5. L. S. Mead, S. J. Arnold, *Trends Ecol. Evol.* **19**, 264 (2004).
6. A. Qvarnstrom, *Trends Ecol. Evol.* **16**, 5 (2001).
7. S. Alonzo, B. Sinervo, *Behav. Ecol. Sociobiol.* **49**, 176 (2001).
8. U. Candolin, *Biol. Rev.* **78**, 575 (2003).
9. A. Janetos, *Behav. Ecol. Sociobiol.* **7**, 107 (1980).
10. R. Gibson, T. Langen, *Trends Ecol. Evol.* **11**, 468 (1996).
11. A. Chaine, thesis, University of California, Santa Cruz (2006).
12. Materials and methods are available as supporting material on *Science* Online.
13. D. Schluter, *Evol. Int. J. Org. Evol.* **42**, 849 (1988).
14. M. Reid, P. Weatherhead, *Anim. Behav.* **40**, 538 (1990).
15. G. Hill, P. Nolan, A. Stoehr, *Behav. Ecol.* **10**, 48 (1999).
16. H. Gibbs, P. Grant, *Nature* **327**, 511 (1987).
17. C. Lessells, P. Boag, *Auk* **104**, 116 (1987).
18. A. Qvarnstrom, T. Part, B. Sheldon, *Nature* **405**, 344 (2000).
19. K. Oh, A. Badyaev, *Proc. R. Soc. London B. Biol. Sci.* **273**, 1913 (2006).
20. R. Lande, S. Arnold, *Evol. Int. J. Org. Evol.* **37**, 1210 (1983).
21. J. Merilä, B. C. Sheldon, L. E. B. Kruuk, *Genetica* **112-113**, 199 (2001).
22. P. R. Grant, B. R. Grant, *Science* **296**, 707 (2002).
23. S. Ellner, *Bull. Math. Biol.* **58**, 103 (1996).
24. M. Kirkpatrick, *Evol. Int. J. Org. Evol.* **36**, 1 (1982).
25. T. Bakker, A. Pomiankowski, *J. Evol. Biol.* **8**, 129 (1995).
26. M. D. Jennions, M. Petrie, *Biol. Rev.* **72**, 283 (1997).

27. We thank J. Bradbury, R. Calsbeek, N. Dominy, R. Montgomerie, and K. Wasson for comments on a draft; B. Sinervo, W. Koenig, A. Ritter, E. Svensson, E. Ferree, D. Shizuka, J. Barna, and T. Price for feedback; J. Barna and volunteers for assisting with data collection; K. Mesa, G. Pogson, M. Burford, C. Fernandez, and J. Thompson for advice on genetics work completed at the University of California, Santa Cruz (UCSC) Molecular Ecology and Evolutionary Genetics (MEEG) facility, and the Pawnee National Grassland and Jones Family for use of their land. Funded by UCSC–Special Research Grant, National Geographic Society, NSF–Doctoral Dissertation Improvement Grant (DDIG) (IBN-0309215), NSF–Graduate Assistantships in Areas of National Need (GAANN), American Museum of Natural History Chapman Fund, Sigma Xi, and the American Ornithologists' Union.

Supporting Online Material

www.sciencemag.org/cgi/content/full/319/5862/459/DC1
Materials and Methods
SOM Text
Figs. S1 to S4
References

13 August 2007; accepted 14 December 2007
10.1126/science.1149167

Control of Genic DNA Methylation by a jmjC Domain–Containing Protein in *Arabidopsis thaliana*

Hidetoshi Saze,* Akiko Shiraishi, Asuka Miura, Tetsuji Kakutani

Differential cytosine methylation of repeats and genes is important for coordination of genome stability and proper gene expression. Through genetic screen of mutants showing ectopic cytosine methylation in a genic region, we identified a jmjC-domain gene, *IBM1 (increase in bonsai methylation 1)*, in *Arabidopsis thaliana*. In addition to the ectopic cytosine methylation, the *ibm1* mutations induced a variety of developmental phenotypes, which depend on methylation of histone H3 at lysine 9. Paradoxically, the developmental phenotypes of the *ibm1* were enhanced by the mutation in the chromatin-remodeling gene *DDM1* (*decrease in DNA methylation 1*), which is necessary for keeping methylation and silencing of repeated heterochromatin loci. Our results demonstrate the importance of chromatin remodeling and histone modifications in the differential epigenetic control of repeats and genes.

Genomes of vertebrates and plants contain a substantial proportion of transposons and repeats (*1*). These potentially deleterious sequences are cytosine-methylated and inactivated (*2*, *3*) to form heterochromatin (*4*, *5*). Methylated heterochromatin, especially when dispersed within gene-rich regions, has the potential to spread by self-reinforcing mechanisms (*6*, *7*) to flanking cellular genes and disrupt their expression. Mechanisms that confine the methylated regions remain enigmatic, despite their importance in maintaining the integrity of large genomes with a high proportion of dispersed transposons. Here, we identify a new pathway that excludes cytosine methylation from genic regions by histone modification and chromatin remodeling, thus ensuring proper plant development.

In the flowering plant *Arabidopsis thaliana*, and in plants in general, cytosine methylation is found in both CG and non-CG contexts. In *Arabidopsis*, methylation at CG sites is maintained by the DNA methyltransferase MET1, whereas methylation at non-CG sites requires the DNA methyltransferase CMT3 (*8*–*12*). Non-CG methylation is also controlled by methylation of histone H3 at lysine 9 (H3mK9) and by the RNA interference (RNAi) machinery (*13*–*16*). DDM1, a chromatin-remodeling adenosine triphosphatase, is involved in maintenance of both CG and non-CG methylation (*17*–*19*). Mutations in *MET1* and *DDM1* also result in a variety of developmental abnormalities by inducing heritable changes in other loci (*8*–*10*, *20*, *21*). One of the *ddm1*-induced abnormalities, called *bonsai* (*bns*), is caused by epigenetic silencing of a gene encoding a homolog of a cell cycle regulator, *APC13* (*22*). The silencing of this gene, *BONSAI* (*BNS*), is associated with spreading of methylated heterochromatin from a flanking LINE retroelement (*22*). This LINE functions as methylated heterochromatin, which has a potential to spread to the flanking *BNS* gene (Fig. 1A).

To explore the mechanisms that exclude genic cytosine methylation in wild-type plants, we used methylation-sensitive restriction enzymes to screen a mutagenized population for individuals with *ibm* (*increase in BONSAI methylation*) phenotype (*23*). One of them, *ibm1*, is described in this report. The *IBM1* gene (*At3g07610*) was identified by a map-based approach (*23*). The original *ibm1-1* mutant has a base substitution causing an amino acid substitution ($Gly^{672} \rightarrow Glu$). We subsequently tested three additional *ibm1* alleles carrying T-DNA insertions and verified that these independent alleles also caused DNA hypermethylation of the *BNS* gene (Fig. 1B).

The *BNS* sequence was hypermethylated in the first generation in which the *ibm1* mutant allele became homozygous (Fig. 1B). This feature was different from the *BNS* hypermethylation in the *ddm1* mutant, which is slow and detectable only after several generations of self-pollination in the mutant background (*22*). Bisulfite sequencing revealed that cytosine methylation occurred at the *BNS* gene in *ibm1* and that non-CG sites are the main targets of the *BNS* methylation (Fig. 1C and table S1). Unlike *ddm1*, the *ibm1* mutation did not affect methylation in repeat

Department of Integrated Genetics, National Institute of Genetics, Yata 1111, Mishima, Shizuoka 411-8540, Japan.

*To whom correspondence should be addressed. E-mail: hsaze@lab.nig.ac.jp