alteration of its poly(A) status (*16*), and we have shown that hunchback mRNA is polyadenylated during development (*29*). Because hunchback mRNA is translationally activated at egg deposition and nanos protein should appear before hunchback protein, a poly(A)-independent mechanism for nanos mRNA might ensure earlier synthesis of this protein.

Polyadenylation of maternal mRNAs is a conserved mechanism of translational activation during oogenesis and early embryogenesis. Therefore, mutations in the pathway might behave as female-sterile mutations, given that maternal mRNAs that require cytoplasmic polyadenylation would not be translationally activated. The isolation and characterization of such *Drosophila* mutants should allow definition of gene products and biochemical pathways that control the initiation of development.

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Trans-Acting Dosage Effects on the Expression of Model Gene Systems in Maize Aneuploids

Mei Guo and James A. Birchler*

The reduction in vigor of aneuploids was classically thought to be due to the imbalance of gene products expressed from the varied chromosome relative to those from the remainder of the genome. In this study, the dosage of chromosomal segments was varied, but the transcript level of most genes encoded therein showed compensation for the number of copies of the gene. Genes whose dosage was not altered were affected by aneuploidy of unlinked chromosomal segments. The phenotypic effects of aneuploidy and of a substantial fraction of quantitative variation are hypothesized to be the consequence of an altered dosage-sensitive regulatory system.

The phenotypic consequences of addition or subtraction of a chromosome relative to the normal genomic complement have been documented in a variety of eukaryotic organisms (*1–3*). The detrimental effects of aneuploidy were considered to be the result of the imbalance of the gene products encoded on the varied chromosome relative to those from the remainder of the genome (*1–3*). This concept was based on the assumption that the amount of gene products increases proportionally as the structural gene dose increases, a concept known as gene dosage effects. However, studies on *Alcohol dehydrogenase 1* (*Adh1*) and *Globulin 1* (*Glb1*) in maize (*Zea mays*) revealed that the expression levels of the ADH1 enzyme and GLB1 protein remained constant for various doses of the long arm of chromosome 1 (1L), where both structural genes reside (*4*, *5*). This lack of a structural gene dosage effect is referred to as dosage compensation. However, when the copy number of the respective structural genes was varied from one to four in a ploidy series, directly proportional expression amounts of *Adh1* and *Glb1* were observed relative to the gene copy number. In addition, most other aneuploids analyzed exhibited trans-acting dosage effects on the expression of genes encoded on unvaried chromosomes (*5*). We used a genetically marked dosage series of 14 chromosomal regions and several specific genes in *Z. mays* to test the effects of segmental ploidy on gene expression. Transcript amounts of the following genes were analyzed: *Adh1* (*6*, *7*), *Alcohol dehydrogenase 2* (*Adh2*) (*7*, *8*); *Glb1* (*9*), *Shrunken 1* (*Sh1*) (*10*), *Sucrose synthase 1* (*Sus1*) (*11*), and *Zein* (22 kD) (*12*).

The genetic system we used to generate the dosage series involved translocations between the supernumerary B chromosome and the normal A chromosomes marked with dominant anthocyanin pigment genes or their transposable element derivatives (*13*). The B chromosome frequently nondisjoins at the second pollen mitosis (*14*), which gives rise to the two sperm in a pollen grain, thus generating zero, one, or two copies of the B chromo-

Division of Biological Sciences, University of Missouri, Columbia, MO 65211, USA.

*To whom correspondence should be addressed.

some in different sperm. We produced a dosage series for a specific A chromosomal segment by crossing a B-A translocation line as the male parent with a normal tester line, which had a recessive marker on the chromosome arm to be analyzed. Dosage classes were identified by following the anthocyanin markers in the embryos and endosperm of the progeny (*15*).

Total RNA from embryo and endosperm tissues harvested 30 days after pollination was extracted (*16*) and subjected to Northern (RNA) analyses (*17*). We examined the expression of the six genes by probing with ^{32}P-labeled antisense RNA. The same blots were then probed with antisense ribosomal RNA (rRNA) (*18*) as a loading control. To confirm that rRNA [encoded on the short arm of chromosome 6 (6S)] expression was not subject to modulation by those chromosomal regions examined, we purified total DNA and RNA from each dosage series. The DNA:rRNA ratios were determined, but no evidence for rRNA effects were found in any of the tested segments (Table 1).

We were able to examine gene expression with various chromosomal doses of the structural genes of *Adh1* (1L), *Glb1* (1L), *Adh2* (4S), and *Sh1* (9S). The B-A translocation line that contains the *Sus1* structural gene is not available in marked form for this type of analysis, and the *Zein* gene used (22 kD) has not been localized. Expression of *Adh1*, *Glb1*, and *Adh2* was relatively constant in the dosage series of the chromosome arms that contain their structural genes (Table 2). This dosage compensation was found in both embryo and endosperm tissues, although *Adh1* showed elevated expression in trisomic embryo tissue. Therefore, dosage compensation seems to be a common phenomenon in whole chromosome arm aneuploids although expression of *Sh1* was correlated with the dosage of 9S, exhibiting a structural gene dosage effect.

In the dosage series of each of the chromosomal regions, we found that transcript amounts of several of the genes were modulated (Table 2). For example, mRNA amounts of *Adh1* in embryo tissues changed in response to the dosage of the 6L, 7L, and 10L regions (Fig. 1). As the dosage of these arms increased, the amounts of mRNA decreased. Other effects, observed as the dosage of 7L increased, were a decrease of *Sus1* in embryo tissues and an increase of *Glb1* and a decrease of *Zein* in endosperm tissues (Fig. 2).

The results suggest that for any one gene, multiple dosage-sensitive modifiers are present in the genome. None of the chromosomal regions affected all of the six genes, suggesting that the effects were exerted through gene-specific, potentially overlapping, systems. In several cases, the dosage effects were not observed throughout the whole dosage series but were absent from either monosomics or trisomics (Table 2). The reasons for the asymmetric responses to ploidy remain unknown.

Dosage-sensitive regulatory effects are tissue specific. The examined genes that were expressed in both embryo and endosperm tissues (*Glb1*, *Adh1*, *Adh2*, and *Sus1*) often showed distinct expression patterns in the two tissues (Fig. 3 and Table 2). The short arm of chromosome 4 inhibited expression of *Sus1* in embryo tissue but enhanced its expression in endosperm. Whether such differential regulation involves the same genetic factors is not known. In embryo tissue, no enhancing effects were seen, whereas in endosperm both enhancement and inhibition were observed. Evolutionarily related genes, such as *Adh1* and *Adh2*, and *Sus1* and *Sh1*, exhibited distinct pat-

Table 1. DNA:rRNA ratios among the dosage series. Total nucleic acid from each set (1 μg) was separated on an agarose gel and stained with ethidium bromide (we tested 1 μg of nucleic acid to show that it fell in the linear response range). Negatives of film (Type 55, Polaroid) were prepared, and the DNA and rRNA bands were scanned with a laser densitometer. DNA:rRNA ratios were then calculated according to the densitometric readings (*17*). CA, chromosome arm.

CA*	Embryo dose			Endosperm dose		
	1	2	3	2	3	4
1Sb	0.76	0.85	0.76	0.89	0.97	0.89
1La	1.02	1.03	1.14	0.90	0.96	0.90
2Sa	1.07	1.06	1.15	0.96	0.91	1.01
3La	1.29	1.28	1.31	1.40	1.43	1.48
4Sa	1.05	1.04	1.04	1.02	1.01	1.01
4Lb	0.40	0.34	0.40	0.57	0.60	0.55
5Sc	0.64	0.65	0.62	0.96	0.92	0.99
5Ld	1.41	1.49	1.44	0.74	0.86	0.79
6Lc	0.57	0.53	0.57	0.58	0.58	0.61
7Lb	1.39	1.30	1.39	1.43	1.12	1.57
8Lc	1.12	1.24	1.18	0.57	0.57	0.58
9Sd	0.69	0.67	0.70	0.80	0.84	0.90
10L19	0.78	0.77	0.81	1.22	1.21	1.33
10L32	0.98	1.10	1.05	1.01	1.00	1.01

*The designation is the chromosome number, followed by S for short or L for long arm. The lowercase letter or number refers to the specific B-A translocation used (*23*). Because the marker systems are present in diverse genetic backgrounds (*13*, *23*), comparisons can only be made among the doses of a particular chromosome arm.

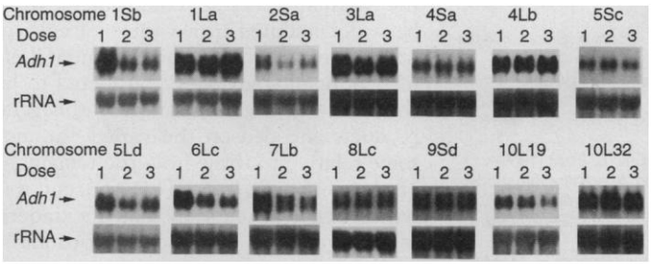

Fig. 1. Expression of *Adh1* in the embryo at various doses of 14 chromosomal regions. The blots were probed with a ^{32}P-labeled antisense RNA of *Adh1* and then reprobed with antisense rRNA (*17*).

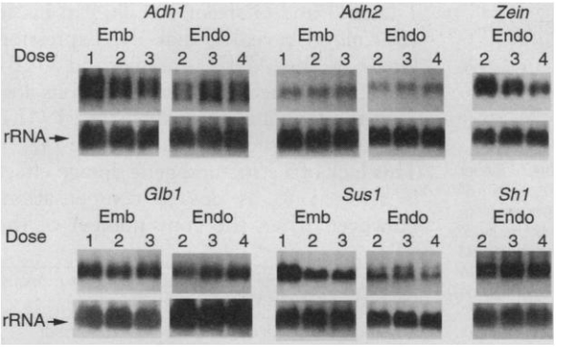

Fig. 2. Expression of the six genes in a dosage series of chromosome arm 7L. Northern blots were first probed with the respective genes and then with rRNA. Emb, embryo; Endo, endosperm.

terns of dosage effects, which suggest that the dosage response of duplicate genes has diverged with their developmental programming.

We have demonstrated a pattern of dosage effects that involves modulation of the expression of all six genes observed. Because only slightly more than half of the maize genome was analyzed, the number of trans-acting effects on any one gene is potentially even greater. Expression of both mRNA and protein from genes varied on whole chromosome arms is often dosage compensated (*4*, *5*, *19*), in contrast with aneuploids immediately around a structural gene where gene dosage effects occur (*20*). The varied chromosomal regions tend to exhibit multiple trans effects (enhancing or inhibiting) on the expression of genes encoded elsewhere. We hypothesize that this spectrum of effects results from altering the dosage of the individual components of the gene regulatory systems that affect each structural gene.

Dosage effects may contribute to the molecular basis of quantitative trait variation. Such variation is often polygenic and exists for many measurable plant characteristics (*21*). The genes that contribute to quantitative variation are of interest because most mutations in general are known to exhibit a dominant or recessive phenotypic pattern in a heterozygote (*22*).

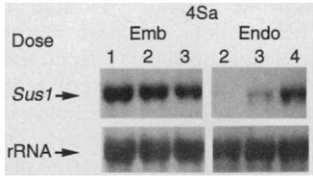

Fig. 3. Tissue-specific modulation on the expression of *Sus1* by chromosome arm 4S. The blots were probed with *Sus1* and then reprobed with rRNA.

Table 2. The mRNA levels in the dosage series relative to the normal euploid (two-dose embryo and three-dose endosperm). When preparing the autoradiographic images for densitometric scanning, we minimized the saturation of the film by controlling the exposure time. The expression levels of various doses relative to that of the euploid were determined by the ratios of respective normalized values (*17*) to that of the euploid (one dose to two doses and three doses to two doses for the embryo; two doses to three doses and four doses to three doses for the endosperm, respectively). The average ratio of three replicates (mean) and the standard error (SE) of each dose (D) are presented. Results in which the chromosomal segment significantly ($P < 0.05$ in t tests) affected RNA amounts (inversely or directly) throughout all ploidies are boxed. Individual trisomics or monosomics that are significantly different from the euploids are shaded. There are 17 total cases of inverse effect and 9 of direct effect. Because of an inverse effect, the lowest reduction in a three-dose embryo is expected to be 67% (2/3) of the normal euploid; the highest expected increase in a one-dose embryo would be 200% (2/1). No trans-acting direct effects were observed in the embryo for these genes. An inverse effect will result in an increase of up to 150% in a two-dose endosperm (3/2) and a reduction to 75% in a four-dose endosperm (3/4). A direct effect will give a reduction to 67% in a two-dose (2/3) and an increase of up to 133% in a four-dose endosperm (4/3).

CA	*Glb1*						*Adh1*						*Adh2*						*Sus1*						*Sh1*			*Zein*		
	Embryo			Endosperm			Embryo			Endosperm			Embryo			Endosperm			Embryo			Endosperm			Endosperm			Endosperm		
	D	Mean	SE	D	Mean	SE	D	Mean	SE	D	Mean	SE	D	Mean	SE	D	Mean	SE	D	Mean	SE	D	Mean	SE	D	Mean	SE	D	Mean	SE
1Sb	1	1.04	0.03	**2**	**0.63**	**0.01**	**1**	**1.72**	**0.03**	**2**	**0.73**	**0.03**	**1**	**1.03**	**0.01**	**2**	**1.13**	**0.02**	**1**	**1.35**	**0.02**	2	0.98	0.01	**2**	**0.72**	**0.02**	2	0.95	0.02
	2	1.00	0.00	**3**	**1.00**	**0.00**	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	**2**	**1.00**	**0.00**	3	1.00	0.00	3	1.00	0.00	3	1.00	0.00
	3	0.97	0.02	**4**	**1.03**	**0.01**	3	0.94	0.03	4	1.12	0.05	3	1.00	0.01	4	1.02	0.01	**3**	**0.88**	**0.02**	4	0.99	0.01	4	1.02	0.04	4	1.03	0.02
1La	1	1.01	0.03	2	0.97	0.02	1	1.23	0.10	2	0.99	0.04	1	1.04	0.04	2	0.97	0.04	**1**	**1.27**	**0.06**	**2**	**0.86**	**0.01**	1	0.91	0.03	2	1.40	0.09
	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	**3**	**1.00**	**0.00**	3	1.00	0.00	3	1.00	0.00
	3	1.03	0.05	4	0.94	0.06	**3**	**1.38**	**0.09**	4	0.99	0.04	3	1.03	0.02	4	0.97	0.02	3	1.01	0.01	**4**	**1.20**	**0.03**	4	1.03	0.02	4	0.91	0.04
2Sa	**1**	**1.38**	**0.07**	2	1.07	0.07	1	0.99	0.03	2	0.97	0.01	1	1.01	0.02	2	0.99	0.01	1	1.00	0.02	2	1.03	0.03	2	0.98	0.02	2	1.03	0.05
	2	**1.00**	**0.00**	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	3	1.00	0.00	3	1.00	0.00
	3	**0.84**	**0.03**	**4**	**0.62**	**0.02**	3	1.02	0.02	4	0.98	0.03	3	1.06	0.04	4	0.98	0.04	**3**	**1.33**	**0.05**	**4**	**1.35**	**0.06**	**4**	**0.82**	**0.01**	4	1.02	0.03
3La	**1**	**1.15**	**0.02**	**2**	**1.37**	**0.06**	1	1.07	0.08	2	1.00	0.06	1	1.00	0.06	**2**	**0.91**	**0.01**	**1**	**1.31**	**0.04**	2	1.00	0.09	2	1.00	0.02	2	0.89	0.03
	2	**1.00**	**0.00**	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	3	1.00	0.00	3	1.00	0.00
	3	**0.68**	**0.05**	4	1.01	0.03	3	1.17	0.04	4	0.94	0.03	3	0.93	0.05	4	0.88	0.03	3	0.88	0.04	4	1.00	0.05	4	0.97	0.02	4	1.02	0.05
4Sa	1	1.04	0.04	2	1.02	0.02	1	1.01	0.03	2	1.13	0.07	1	0.96	0.04	2	0.99	0.02	**1**	**1.14**	**0.01**	**2**	**0.58**	**0.06**	**2**	**0.79**	**0.02**	**2**	**0.53**	**0.03**
	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	**2**	**1.00**	**0.00**	**3**	**1.00**	**0.00**	**3**	**1.00**	**0.00**	**3**	**1.00**	**0.00**
	3	0.84	0.06	**4**	**0.83**	**0.00**	3	1.03	0.06	4	0.97	0.02	3	1.03	0.06	4	1.05	0.03	**3**	**0.89**	**0.02**	**4**	**2.46**	**0.15**	**4**	**1.17**	**0.01**	**4**	**1.20**	**0.01**
4Lb	1	1.03	0.03	**2**	**0.60**	**0.01**	1	1.04	0.12	2	0.77	0.07	1	1.07	0.06	2	1.00	0.02	1	1.01	0.03	**2**	**0.82**	**0.01**	**2**	**0.66**	**0.03**	2	0.96	0.03
	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	3	1.00	0.00	3	1.00	0.00
	3	1.04	0.04	4	0.97	0.03	**3**	**1.12**	**0.03**	**4**	**0.85**	**0.03**	3	1.03	0.06	4	1.01	0.02	3	1.13	0.04	**4**	**0.83**	**0.03**	4	0.98	0.04	4	0.98	0.02
5Sc	**1**	**1.43**	**0.03**	**2**	**0.47**	**0.04**	1	0.95	0.04	2	1.09	0.06	1	1.00	0.01	2	0.99	0.03	**1**	**0.72**	**0.05**	2	1.07	0.09	**2**	**1.14**	**0.02**	2	0.87	0.04
	2	**1.00**	**0.00**	**3**	**1.00**	**0.00**	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	**3**	**1.00**	**0.00**	3	1.00	0.00
	3	**0.76**	**0.04**	**4**	**1.30**	**0.02**	3	0.96	0.05	4	0.98	0.02	3	0.98	0.01	4	1.01	0.03	**3**	**0.70**	**0.04**	4	1.06	0.04	**4**	**0.88**	**0.02**	4	0.97	0.05
5Ld	**1**	**1.35**	**0.05**	**2**	**1.66**	**0.04**	1	1.15	0.04	2	0.96	0.04	1	1.08	0.01	2	0.97	0.05	**1**	**1.93**	**0.12**	2	1.70	0.19	2	0.97	0.04	**2**	**1.42**	**0.06**
	2	1.00	0.00	**3**	**1.00**	**0.00**	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	**2**	**1.00**	**0.00**	3	1.00	0.00	3	1.00	0.00	3	1.00	0.00
	3	1.01	0.04	**4**	**0.90**	**0.00**	3	1.03	0.03	**4**	**0.91**	**0.01**	3	1.06	0.04	4	1.03	0.07	**3**	**0.82**	**0.02**	**4**	**0.75**	**0.02**	4	1.03	0.04	4	1.02	0.04
6Lc	**1**	**1.68**	**0.09**	2	0.95	0.05	**1**	**1.76**	**0.10**	**2**	**0.86**	**0.03**	1	0.91	0.02	2	1.01	0.01	**1**	**1.47**	**0.01**	2	1.00	0.07	2	0.96	0.02	**2**	**0.78**	**0.02**
	2	**1.00**	**0.00**	3	1.00	0.00	**2**	**1.00**	**0.00**	**3**	**1.00**	**0.00**	2	1.00	0.00	3	1.00	0.00	**2**	**1.00**	**0.00**	3	1.00	0.00	3	1.00	0.00	3	1.00	0.00
	3	**0.64**	**0.03**	4	0.99	0.03	**3**	**0.79**	**0.06**	**4**	**1.12**	**0.03**	3	1.01	0.06	4	1.10	0.03	**3**	**0.91**	**0.00**	**4**	**1.63**	**0.02**	4	1.01	0.03	4	1.09	0.05
7Lb	1	1.00	0.02	**2**	**0.61**	**0.01**	**1**	**2.17**	**0.17**	**2**	**0.75**	**0.01**	1	0.89	0.05	2	0.87	0.05	**1**	**1.81**	**0.03**	2	1.01	0.02	2	0.97	0.05	**2**	**1.38**	**0.09**
	2	1.00	0.00	**3**	**1.00**	**0.00**	**2**	**1.00**	**0.00**	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	**2**	**1.00**	**0.00**	3	1.00	0.00	3	1.00	0.00	**3**	**1.00**	**0.00**
	3	0.99	0.02	**4**	**1.26**	**0.05**	**3**	**0.65**	**0.06**	4	0.96	0.02	**3**	**1.25**	**0.03**	4	1.02	0.05	**3**	**0.85**	**0.02**	4	0.95	0.04	**4**	**1.06**	**0.01**	**4**	**0.70**	**0.03**
8Lc	1	0.92	0.02	2	1.04	0.05	1	0.96	0.01	2	1.21	0.07	1	1.01	0.04	**2**	**0.91**	**0.01**	1	1.02	0.05	2	1.07	0.04	2	1.00	0.15	2	1.01	0.05
	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	3	1.00	0.00	3	1.00	0.00
	3	0.99	0.03	4	0.90	0.06	3	1.10	0.04	4	0.91	0.02	3	1.04	0.04	4	1.03	0.12	3	1.01	0.03	4	0.99	0.02	4	0.93	0.08	4	0.98	0.01
9Sd	1	0.89	0.07	**2**	**1.43**	**0.05**	1	1.00	0.02	**2**	**1.13**	**0.02**	1	0.97	0.03	2	1.01	0.07	1	0.96	0.05	**2**	**1.40**	**0.08**	**2**	**0.63**	**0.08**	2	0.91	0.05
	2	1.00	0.00	**3**	**1.00**	**0.00**	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	**3**	**1.00**	**0.00**	3	1.00	0.00
	3	1.03	0.04	**4**	**0.82**	**0.02**	3	0.99	0.03	**4**	**1.28**	**0.02**	**3**	**0.80**	**0.03**	4	1.03	0.07	3	0.97	0.04	4	0.84	0.04	**4**	**1.57**	**0.12**	4	1.02	0.02
10L19	1	1.10	0.03	2	0.92	0.03	**1**	**1.18**	**0.02**	2	0.97	0.03	1	1.00	0.02	2	0.92	0.03	**1**	**1.16**	**0.04**	2	0.99	0.05	2	0.96	0.06	2	0.96	0.03
	2	1.00	0.00	3	1.00	0.00	**2**	**1.00**	**0.00**	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	**2**	**1.00**	**0.00**	3	1.00	0.00	3	1.00	0.00	3	1.00	0.00
	3	0.98	0.01	4	0.93	0.03	**3**	**0.65**	**0.02**	4	1.03	0.03	3	1.01	0.04	4	0.97	0.02	**3**	**0.67**	**0.02**	**4**	**1.25**	**0.03**	4	1.16	0.09	4	0.96	0.01
10L32	1	1.04	0.03	2	0.99	0.03	1	0.93	0.04	**2**	**0.84**	**0.02**	1	0.86	0.07	2	1.01	0.01	**1**	**1.15**	**0.01**	**2**	**0.65**	**0.02**	**2**	**1.18**	**0.05**	2	1.00	0.02
	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	2	1.00	0.00	3	1.00	0.00	3	1.00	0.00	3	1.00	0.00
	3	0.89	0.01	4	0.98	0.04	3	0.94	0.04	4	1.14	0.04	3	1.04	0.06	4	1.02	0.07	3	1.02	0.02	4	0.99	0.03	4	0.83	0.02	4	0.98	0.03

This fact suggests that a large reduction of most of the relevant gene products is required to produce an abnormal phenotype. In contrast, most quantitative variation exhibits some degree of additive inheritance (*21*). Allelic variation at the loci responsible for the dosage effects described here would be additive. The polygenic and additive nature of both phenomena suggests a relation between the two. Thus, it is likely that a substantial fraction of quantitative variation is the result of polymorphism at the loci responsible for the trans-acting effects.

The overall prevalence of dosage effects on different classes of genes also has implications for the molecular basis of aneuploid syndromes. Monosomics are typically less vigorous than trisomics, which are in turn less vigorous than euploids (*1–3*). This observation correlates with the generalized lowest levels of gene expression found in the respective chromosomal configurations in our study. In other words, the greatest reductions were found in monosomics, and lesser reductions were in trisomics, as compared with the diploids. The finding of a greater set of modulations in the aneuploid series, as opposed to the ploidy series (*5*), suggested that the classically defined imbalance associated with aneuploidy (*1–3*) is a reflection of the underlying regulatory system. We suggest that the reduction of vigor in monosomics and trisomics results from the limiting effects of many gene products encoded on the respective chromosome as well as those from throughout the genome.

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15. In the male parent, the A segment of the B-A chromosome is marked by a dominant allele of an anthocyanin gene, and the normal A chromosome from the female tester line carries a recessive allele. The two unequivalent sperm produced from the nondisjunction of the B-A chromosome could fertilize the egg cell or the two polar nuclei. Fertilization of the egg cell with the zero-dose gamete and of the polar nuclei with the two-dose gamete gave rise to embryos of one dose and endosperm of four doses of the corresponding A chromosomal segment. This class was identified as kernels with colorless embryos and colored endosperm. The opposite fertilization event produced three-dose embryos and two-dose endosperm, which could be identified as colored embryos and colorless endosperm. Fertilization by equivalent gametes (each carrying one dose of the B-A chromosome) formed by disjunction of the B-A chromosome resulted in normal euploid kernels with two-dose embryos and three-dose endosperm, which could be recognized as those with both pigmented embryos and endosperm.
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17. Northern analysis was performed as described [J. A. Birchler and J. C. Hiebert, *Genetics* **122**, 129 (1989)]. For each specific gene, triplicate loadings of each dosage series were analyzed in each blot. The autoradiographic images were quantitated by laser densitometry. We normalized the specific mRNA quantity by calculating the ratio of mRNA to rRNA of the densitometric readings. The normalized values (mRNA:rRNA ratio) of the three replicates were used for statistical analyses.
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Binding and Stimulation of HIV-1 Integrase by a Human Homolog of Yeast Transcription Factor SNF5

Ganjam V. Kalpana, Shana Marmon, Weidong Wang, Gerald R. Crabtree, Stephen P. Goff*

Upon entry into a host cell, retroviruses direct the reverse transcription of the viral RNA genome and the establishment of an integrated proviral DNA. The retroviral integrase protein (IN) is responsible for the insertion of the viral DNA into host chromosomal targets. The two-hybrid system was used to identify a human gene product that binds tightly to the human immunodeficiency virus–type 1 (HIV-1) integrase in vitro and stimulates its DNA-joining activity. The sequence of the gene suggests that the protein is a human homolog of yeast SNF5, a transcriptional activator required for high-level expression of many genes. The gene, termed *INI1* (for integrase interactor 1), may encode a nuclear factor that promotes integration and targets incoming viral DNA to active genes.

The retroviral integrase enzyme catalyzes two specific reactions: (i) cleavage of the 3′-termini of the viral DNA to produce recessed 3′-OH ends, and (ii) joining of the two newly generated 3′-termini to the 5′-phosphates on each strand of the target sequence in a concerted strand-transfer reaction (*1*). Although recombinant integrase preparations can carry out all the steps known to be required for processing and joining of the viral DNA, there are indications of the involvement of additional factors in integration. For example, the isolated proteins show only very low specific activities for cutting and joining of DNA in vitro (*2*). Furthermore, for some viruses, joining reactions carried out with oligonucleotide substrates result in the transfer of only one 3′-OH to the target DNA, yielding a Y structure, rather than the concerted transfer of two 3′-OH termini to the target. It has been proposed that the viral DNA is restrained from integration into itself (autointegration) by a proposed inhibitor that can be removed by high concentrations of salt (*3*). Finally, integration in vivo is thought to be biased toward targets near transcriptionally active genes or open chromatin (*4*). Although the mechanism for such bias is not clear, transcription factors that bind DNA in a site-specific manner are thought to mediate target site selection by related retrotransposable elements (*5*).

We used the two-hybrid system (*6*) to identify host proteins that bind to the HIV-1 IN. Approximately 10^6 complementary DNAs (cDNAs) of the HL60 macrophage-monocytic cell line were expressed as GAL4AC (activation domain) fusions and tested for coactivation of a reporter gene together with a GAL4DB (DNA binding) IN fusion (*7*). Three iso-

G. V. Kalpana, S. Marmon, S. P. Goff, Howard Hughes Medical Institute, Department of Biochemistry and Molecular Biophysics, Columbia University, College of Physicians and Surgeons, 630 West 168th Street, New York, NY 10032, USA.
W. Wang and G. R. Crabtree, Howard Hughes Medical Institute, Department of Developmental Biology, Beckman Center B207, Stanford University, Stanford, CA 94305, USA.

*To whom correspondence should be addressed.