Genet. Res., Camb. (1991), **57**, *pp.* 55–59 *Printed in Great Britain*

Partial complementation by murine *t* haplotypes: deficit of males among t^6/t^{w5} double heterozygotes and correlation with transmission-ratio distortion

THOMAS R. KING*

Laboratory of Genetics and McArdle Laboratory for Cancer Research, University of Wisconsin-Madison, Madison, Wisconsin 53706

(*Received 8 May 1990 and in revised form 18 June 1990*)

Summary

To evaluate whether sex reversal contributes to sex-ratio imbalance among t^6/t^{w5} double heterozygotes, the cross performed by K. B. Bechtol (*Genetical Research* **39**, 1982, 79–84), $T/t^6 \times T/t^{w5}$, was repeated. Significantly more normal-tailed (t^6/t^{w5}) females than males were recovered. By contrast, sex ratios were normal among tailless progeny resulting from this cross and among all classes produced by control crosses. Hybridization of a Y-specific DNA probe with genomic DNA from phenotypic females revealed no XY, sex-reversed males. On the genetic backgrounds that generated only moderate transmission distortion of t^{w5} (81–85%), the overall viability of the doubly heterozygous progeny was only 50% and the sex-ratio skew among this class was strong. However, on a genetic background that displayed extreme t^{w5} transmission (99%), embryonic viability was more than 80% and the sex-ratio imbalance was weak.

1. Introduction

In wild mouse populations, the proximal third of chromosome *17* (Chr *17*) is commonly found in variant forms, *t* haplotypes, that control several striking genetic effects. These include: interaction with brachyury (*T*) to produce taillessness in T/t heterozygotes; altered transmission ratio of *t* chromosomes from heterozygous males; and crossover suppression over the entire *t* region (reviewed by Silver, 1985). In addition, at least 14 embryonic lethal (*tcl*) complementation groups are associated with different *t* haplotypes, although not all *t* haplotypes carry recessive lethal mutations (Bennett, 1975; Magnuson, 1983; Klein, Sipos & Figueroa, 1984). Utilizing free recombination between complementing *t* haplotypes, Artzt and coworkers (Artzt, McCormick & Bennett, 1982; Artzt, 1984) and Pla & Condamine (1984) showed that tcl^t mutations belonging to different complementation groups map to independent loci throughout the *t* region.

Only partial complementation has been observed between *t* haplotypes from different complementation groups, denoted here as t^a and t^b. Often, t^a/t^b heterozygotes from $T/t^a \times T/t^b$ intercrosses show reduced viability (18% to 85%) relative to their tailless (T/t) sibs (Dunn & Gluecksohn-Shoenheimer, 1943; Dunn, 1956; Smith, 1956; Silagi, 1962; Bennett, 1975; Bechtol, 1982; Shin *et al.* 1983; Mains, 1986). In addition, males doubly heterozygous for complementing *t* haplotypes are sterile (Lyon, 1986). In at least some cases, t^a/t^b males display mortality in excess of t^a/t^b females (Dunn & Gluecksohn-Shoenheimer, 1943; Bechtol, 1982).

One model to explain the skewed sex ratios (in favour of females) that have been observed among mice doubly heterozygous for certain partially-complementing *t* haplotypes would posit that a *t*-region gene with a role in primary sex determination is altered in each member of these pairs of *t* haplotypes. Such alterations could disrupt the normal interaction of this gene or gene product with signals initiated by the Y-linked testis-determining locus (*Tdy*), resulting in a high frequency of XY females among t^a/t^b double heterozygotes. Indeed, Eicher & Washburn (1983, 1986, 1989, 1990) have identified a gene or gene complex (*Tas*) delimited by the hairpin tail (T^{hp}) and T-Orleans (T^{Orl}) deletions on Chr *17* that affects primary sex determination. They observe that C57BL/6J mice cogenic for $T^{hp}/+$ or $T^{Orl}/+$ and carrying the Y-linked testis-determining locus from strain AKR/J (Tdy^a) either display hermaphroditic gonads or are completely sex reversed. In an alternative model, sex-ratio imbalance among t^a/t^b double heterozygotes could be due to factors independent of *Tas* and *Tdy*. For example, additive sublethal effects of these *t* haplotypes might slow the growth of

* Present address: The Jackson Laboratory, 600 Main Street, Bar Harbor, ME 04609, USA.

t^a/t^b embryos; slow embryonic growth (e.g. from haploinsufficiency) has been postulated to be responsible for XY sex reversal (Cattanach, Rasberry & Beechey, 1988; Mittwoch, 1989). Finally, instead of sex reversal, the observed excess of females might involve the differential viability of male versus female embryos.

To evaluate whether sex reversal contributes to skewed sex ratios among t^6/t^{w5} double heterozygotes, a cross similar to that performed by Bechtol (1982) has been repeated, and genomic DNA from the resulting t^6/t^{w5} females has been tested for hybridization with a Y-specific DNA probe.

2. Materials and methods

(i) *Mice*

T/t^6 mice, obtained from The Jackson Laboratory (Bar Harbor, ME), were crossed with brachyury, tufted (Lyon, 1956) $T\,tf/+tf$ partners from the segregating inbred strain BTBR (obtained from J.-L. Guénet, Institut Pasteur, Paris). The t^6 haplotype has been maintained in subsequent generations by intercrossing tailless, non-tufted brothers and sisters. The $T\,tf/t^6$ and $T\,tf/+tf$ mice used in experiments described here were the sister progeny from F_1 and F_2 females that were backcrossed to short-tailed $(T/+)$ BTBR males. $T\,tf/t^{w5}$ mice were obtained from V. C. Bode (Kansas State University, Manhattan, KS). These mice were backcrossed (N_2) to strain C57BL/6J, followed by brother–sister intercrossing for eight to nine generations. C57BL/6J mice cogenic for the brachyury allele T-Oak Ridge 4 (T^{4Or}; Bennett *et al.* 1975), B6-T^{4Or}, or the Y chromosome from strain AKR/J, B6.AKR-Y, were obtained from E. M. Eicher (The Jackson Laboratory, Bar Harbor, ME). $T^{4Or}/+$, XY^{AKR} males were produced by mating B6-T^{4Or} females (N_{18}) and B6.AKR-Y males (N_{18}) and then backcrossing to C57BL/6J females. Mice carrying T^{hp} (Johnson, 1974; 1975) were obtained from L. M. Silver (Princeton University, Princeton, NJ). The male $T^{hp}/+$ carriers used in these experimemts were N_9 on the BTBR background. T/t^{w5} males carrying the Y chromosome from strain AKR (Y^{AKR}) were produced by crossing females from the $T\,tf/t^{w5}$ line with $T^{4Or}/+$, XY^{AKR} or $T^{hp}/+$, XY^{AKR} males.

(ii) *Southern blot analysis*

DNA for Southern blot analysis was prepared from whole blood by the method of Phillips & Nadeau (1984), digested with *Eco*RI or *Pst*I and electrophoresed through 0.8% or 1.0% agarose. Gels were stained with ethidium bromide and photographed. As a positive control, DNA from at least one male was included on every gel and was loaded into several lanes in diminishing amounts, from one-half to one-tenth of a single DNA preparation. Only those experimental lanes that showed at least as much ethidium-bromide staining as positive-control lanes were counted. Gels were depurinated with 0·25 M-HCl and transferred to Zetaprobe (Bio-Rad) filters with 0·4 M-sodium hydroxide, according to the manufacturer's instructions. Filters were prehybridized for 2–4 h at 65 °C in 0·25 M sodium phosphate, pH 7·2/5% sodium dodecyl sulphate/1 mM-EDTA/10% (wt/vol) PEG, molecular weight 6000–8000/sonicated calf thymus DNA (100 μm/ml). Whole plasmid or purified insert from pYB10 (Eicher *et al.* 1989), donated by E. M. Eicher, was labelled with [α-^{32}P]dCTP by nick translation (Rigby *et al.* 1977) or by random primer extension (Feinberg & Vogelstein, 1984). Hybridizations were carried out for 16 to 18 h at 65 °C. Filters were washed to high stringency (final wash in 0·04 M sodium phosphate, 1% sodium dodecyl sulphate at 60 °C) with gentle agitation. Washed filters were exposed for 48–72 h to Kodak XAR-5 film with intensifying screens.

(iii) *Statistical analysis*

Standard error (S.E.) was estimated as described by Bhattacharyya & Johnson (1977, p. 242). Statistical inferences concerning comparisons between population proportions were made according to the method described on pp. 308–312.

3. Results

(i) *Incomplete complementation between* t^6 *and* t^{w5} *confirmed*

Mice were mated as shown in Table 1. Individual $T\,tf/t^{w5}$ males were simultaneously bred with experimental $T\,tf/t^6$ and control $T\,tf/+tf$ females throughout the study. The sex and tail phenotypes of the offspring were recorded one to two days after birth and were reconfirmed at weaning. While the full penetrance of T was not verified by progeny testing, tufted was scored among the progeny of the $T\,tf/+tf$ (control) females so that any non-penetrant $T\,tf/+tf$ mice could be detected among apparently normal-tailed progeny. No phenotypic misclassifications were discovered.

Among the progeny of the control $T\,tf/+tf$ females, sex ratios were not significantly different from 1 female:1 male (in the worst case, that of the short-tailed class, $P > 0{\cdot}1$). Thus, X- and Y-bearing sperm appeared to be transmitted with equal frequency and male and female zygotes appeared to survive equally well. In addition, the set of $T\,tf/t^{w5}$ males was shown to transmit the t^{w5}-bearing chromosome 85% of the time (see Table 1).When these same males were crossed to $T\,tf/t^6$ (experimental) females, only about 50% of the expected t^6/t^{w5} progeny were observed, based on 85% transmission of t^{w5}. Furthermore, the observed sex-ratio – 3 females:2 males – was significantly different from 1:1 ($P \approx 0{\cdot}0015$). The fre-

https://doi.org/10.1017/S0016672300029049 Published online by Cambridge University Press

Table 1. *The tail phenotype and sex of offspring from* T tf/+tf × T tf/t^{w5} *and* T tf/t^6 × T tf/t^{w5}

Cross	*T tf/+tf* ♀						×	*T/tf/t^{w5}* ♂	×	*T tf/t^6* ♀			
Progeny													
Tail (*Genotype*)	Tailless (T/t^{w5})		Short ($T/+$)		Normal ($+/t^{w5}$)			Designation		Tailless (T/t^{w5}; T/t^6)		Normal (t^6/t^{w5})	
Sex	♀	♂	♀	♂	♀	♂				♀	♂	♀	♂
Number	42	21	7	4	34	30		*a*		67	45	26	18
	4	7	2	0	11	9		*b*		9	6	4	2
	16	13	3	2	20	14		*c*		18	13	11	8
	25	27	6	4	36	29		*d*		50	36	17	8
	8	9	3	0	7	9		*e*		21	21	16	5
	10	11	1	2	9	12		*f*		23	39	21	11
	39	36	6	6	24	32		*g*		70	60	30	24
	16	21	6	3	26	27		*h*		22	26	10	11
	160	145	34	21	167	162				280	246	135	87
	305		55		329		= 689		748 =	526		222	
Frequency of males	0·48 ± 0·03		0·38 ± 0·07		0·49 ± 0·03					0·47 ± 0·02		0·39 ± 0·03	
Transmission frequency of t^{w5}	$\frac{305+329}{305+329+2(55)} = 0{\cdot}85$							Expected frequencies based on t^{w5} transmission of 0·85		0·5405		0·4594	
								Expected number of progeny		526		447	
								Viability rates		Total 0·50 ± 0·02			
										Females 0·60 ± 0·03			
										Males 0·39 ± 0·03			

Progeny listed along the same row were sired by the same *T tf/t^{w5}* male. For determining the viability rates for t^6/t^{w5} mice, the expected frequency and number of t^6/t^{w5} mice (assuming full viability of the tailless class and a transmission frequency of 0·85 for t^{w5}) was calculated by assuming the number of T/t^6 and T/T progeny to be equal and the number of T/t^{w5} and t^6/t^{w5} progeny to be equal, since there is no transmission-ratio distortion from the female parent. Frequencies shown are ± 1 S.E.

quency of males among the t^6/t^{w5} mice was also significantly different from the frequency of males observed among tailless siblings ($P \approx 0{\cdot}03$) and from that observed among the *t*-bearing progeny of control *T tf/+tf* mothers ($P \approx 0{\cdot}009$). These results support the male-deficit effect observed by Bechtol (1982), but show a significantly less dramatic sex-ratio skewing than the 3:1 ratio she reported ($P \approx 0{\cdot}025$).

(ii) *Sex-ratio skewing among* t^6/t^{w5} *animals is not enhanced by* Y^{AKR}

The model presented by Eicher & Washburn (1986) for the genetic control of primary sex determination suggests that the Tdy^a allele encoded on the Y chromosome of strain AKR may initiate the testis-determining pathway later than the Tdy^b allele carried by strain C57BL/6J. Thus, the marked deficiency in male progeny reported by Bechtol (1982), compared with the results in Table 1, might be owing to the particular *Tdy* allele involved; the origin of the Y carried by the T/t^{w5} males used by Bechtol is unknown. Complementation and control crosses were therefore repeated with T/t^{w5} males known to carry the Y^{AKR} chromosome; these results are presented in Table 2. While more t^6/t^{w5} females than males were again produced, the skewing is less extreme than for the cross described in Table 1. The frequency of males among the t^6/t^{w5} mice is not significantly different from 0·5 ($P \approx 0{\cdot}19$), or from the frequency of males observed among tailless siblings ($P \approx 0{\cdot}16$), or from that observed among the *t*-bearing progeny of (control) *T tf/+tf* mothers ($P \approx 0{\cdot}08$). Interestingly, t^{w5} transmission from males in the cross shown in Table 2 was significantly higher (99%) than from males in the cross shown in Table 1 (85%) ($P < 0{\cdot}0001$). In addition, the viability of t^6/t^{w5} zygotes produced in the second cross (Table 2; ~ 80%, overall) was significantly elevated ($P < 0{\cdot}0001$), compared with that observed in the first cross (Table 1; ~ 50% viability, overall).

(iii) *Probing for Y-specific sequences among female* t^6/t^{w5} *mice*

Two hundred and twenty-nine t^6/t^{w5} females produced in experimental crosses (134 of 135 from the cross shown in Table 1, 95 of 101 from the cross shown in Table 2) and 23 of their tailless female littermates (all from Table 1) were tested by Southern blot analysis for the presence of a Y-specific sequence (Eicher *et al.* 1989). While DNA from their fathers and brothers

https://doi.org/10.1017/S0016672300029049 Published online by Cambridge University Press

Table 2. *The tail phenotype and sex of offspring from* T tf/+tf × T/t^{w5}, *XY*^{AKR} *and* T tf/t^{6} × T/t^{w5}, *XY*^{AKR}

Cross	*T tf/+tf* ♀				×	*T/t^{w5}* ♂	×	*T tf/t^{6}* ♀			
Progeny											
Tail (*Genotype*)	Tailless (*T/t^{w5}*)		Short (*T/+*)		Normal (*+/t^{w5}*)		Designation	Tailless (*T/t^{w5}*; *T/t^{6}*)		Normal (*t^{6}/t^{w5}*)	
Sex	♀	♂	♀	♂	♀	♂		♀	♂	♀	♂
Number	31	34	0	0	29	30	*i*	22	26	22	24
	15	16	0	0	9	16	*j*	11	12	15	11
	36	41	0	1	47	41	*k*	30	39	11	8
	14	21	0	0	14	19	*l*	9	11	33	29
	4	5	0	0	8	7	*m*	25	17	16	12
	6	5	0	0	10	6	*n*	2	2	1	0
	7	7	0	1	5	5	*o*	3	1	1	0
	4	3	0	0	9	7	*p*	10	6	8	4
	117	132	0	2	131	131		112	114	101	83
	249		2		262	= 513		410 = 226		184	
Frequency of males	0·53 ± 0·03				0·50 ± 0·03			0·50 ± 0·03		0·45 ± 0·04	
Transmission frequency of *t^{w5}*	$\frac{249+262}{249+262+2(2)} = 0.99$						Expected frequencies based on *t^{w5}* transmission of 0·99	0·5019		0·4981	
							Expected number of progeny	226		224	
							Viability rates	Total 0·82 ± 0·03			
								Females 0·92 ± 0·03			
								Males 0·74 ± 0·03			

Progeny listed along the same row were sired by the same *T tf/t^{w5}* male. For determining the viability rates for *t^{6}/t^{w5}* mice, the expected number of *t^{6}/t^{w5}* mice (assuming full viability of the tailless class and a transmission frequency of 0·99 for *t^{w5}*) was calculated by assuming the number of *T/t^{6}* and *T/T* progeny to be equal and the number of *T/t^{w5}* and *t^{6}/t^{w5}* progeny to be equal, since there is no transmission-ratio distortion from the female parent. Frequencies shown are ± 1 S.E. *T/t^{w5}* males *i-m* carried *T^{4Or}* − +, males *n–p* carried *T^{hp}–tf* (see Materials and methods).

hybridized strongly with labelled probe, DNA from all phenotypic females gave no signal. Thus, the sex-ratio skew demonstrated in these crosses is not due to the classification of XY, sex-reversed animals as females.

4. Discussion

Clearly, the sex-ratio imbalance reported here for *t^{6}/t^{w5}* double heterozygotes was not caused by sex-reversed XY animals. However, the mice bred by Bechtol (1982) may have included alleles at *Tdy* and/or other background loci that increased the penetrance of XY sex reversal in *t^{w5}/t^{6}* double heterozygotes. (Because the *t* haplotypes used by Bechtol were not on standard backgrounds, it is not possible to duplicate her experiment precisely and test directly for sex reversal under those conditions.)

It is intriguing that the *T/t^{w5}* males described in Table 2 exhibited a dramatically higher *t^{w5}*-transmission frequency than did the males shown in Table 1. Gummere and coworkers (1986) have described a similar variability in transmission-distortion ratio between two inbred strains that carry *t^{12}*. These authors attribute this effect to genetic differences both on Chr *17* and in the background. Control crosses have shown that the difference in *t^{w5}* transmission between the males in Table 1 versus the males in Table 2 cannot be entirely owing to differences between their Y chromosomes. *T^{4Or}/t^{w5}* males, produced by the reciprocal cross – female *T^{4Or}/+* (from the cross B6-*T^{4Or}* × B6.AKR-Y, as in Materials and methods) × male *T tf/t^{w5}* – transmit *t^{w5}* at 100% (based on 141 sampled gametes).

Among *t^{6}/t^{w5}* progeny sired by males that displayed 99% transmission of *t^{w5}* (Table 2), the viability was 80% and the sex ratio was only slightly skewed (45% males). By contrast, sires that exhibited modest transmission distortion (81%, see Bechtol, 1982; 85%, see Table 1), produced lower *t^{6}/t^{w5}* viability (52%, Bechtol, 1982; 50%, Table 1) and more dramatic sex-ratio skewing (25% males, Bechtol, 1982; 39% males, Table 1). Again, these differences are likely to be due to the genetic backgrounds of the sires since the females used in the crosses presented here were very similar (N_2 cousins). Co-modulation of viability rate, sex-ratio imbalance, and transmission-ratio distortion when genetic backgrounds were altered could indicate a shared mechanism underlying these properties of *t* haplotypes. Alternatively, an entirely different set of genes could be involved.

In any case, the sex-related mortality of certain *t^{a}/t^{b}*

https://doi.org/10.1017/S0016672300029049 Published online by Cambridge University Press

zygotes, reported first by Dunn & Gluecksohn-Schoenheimer (1943), later by Bechtol (1982), and reaffirmed here, remains a puzzling feature of *t*-haplotype genetics. Perhaps the cause of this phenomenon will be elucidated only when the gene products associated with the transmission-ratio distortion phenotype (see Lyon, 1984, 1987; Schimenti *et al.* 1988; Bullard & Schimenti, 1990) become defined, and interactions between them can be studied.

Sincere thanks to Karina Burger and Dan Harrington for technical assistance; William F. Dove, Amy R. Moser and Alexandra Shedlovsky for continuing valuable discussions and criticism; and Eva M. Eicher and Linda L. Washburn for helpful comments concerning this manuscript. This paper is contribution number 3102 from the Laboratory of Genetics, University of Wisconsin–Madison. This work was supported by Predoctoral Training Grants GM-07133 and CA-09135, Program-Project Grant CA-23076, and Core Grant CA-07175, all from the National Institutes of Health.

References

Artzt, K. (1984). Gene mapping within the *T/t* complex of the mouse. III. *t*-lethal genes are arranged in three clusters on chromosome 17. *Cell* **39**, 565–572.

Artzt, K., McCormick, P. & Bennett, D. (1982). Gene mapping within the *T/t* complex of the mouse. I. *t*-lethal genes are nonallelic. *Cell* **28**, 463–470.

Bechtol, K. B. (1982). Lethality of heterozygotes between *t*-haplotype complementation groups of mouse: sex-related effect on lethality of t^6/t^{w5} heterozygotes. *Genetical Research* **39**, 79–84.

Bennett, D. (1975). The *T* complex of the mouse. *Cell* **6**, 441–454.

Bennett, D., Dunn, L. C., Spiegelman, K., Artzt, K., Cookingham, J. & Schermerhorn, E. (1975). Observations on a set of radiation induced dominant T-like mutations in the mouse. *Genetical Research* **26**, 95–108.

Bhattacharyya, G. K. & Johnson, R. A. (1977). In *Statistical Concepts and Methods*. Pp. 249, 308–312. John Wiley & Sons, New York.

Bullard, D. C. & Schimenti, J. C. (1990). Molecular cloning and genetic mapping of the *t complex responder* candidate gene family. *Genetics* **124**, 957–966.

Cattanach, B. M., Rasberry, C. & Beechley, C. V. (1988). Sex reversal and retardation of embryonic development. *Mouse News Letter* **82**, 94.

Dunn, L. C. (1956). Analysis of a complex gene in the house mouse. *Cold Spring Harbor Symposia on Quantitative Biology* **21**, 187–195.

Dunn, L. C. & Gluecksohn-Schoenheimer, S. (1943). Tests for recombination amongst three mutations in the house mouse. *Genetics* **28**, 29–40.

Eicher, E. M., Hutchinson, K. W., Phillips, S. J., Tucker, P. K. & Lee, B. K. (1989). A repeated segment on the mouse *Y* chromosome is composed of retroviral-related, *Y* enriched and *Y*-specific sequences. *Genetics* **122**, 181–192.

Eicher, E. M. & Washburn, L. L. (1986). Genetic control of primary sex determination in mice. *Annual Review of Genetics* **20**, 327–360.

Feinberg, A. P. & Vogelstein, B. (1984). A technique for radiolabelling endonuclease fragments to high specific activity. *Analytical Biochemistry* **137**, 266–267.

Gummere, G. R., McCormick, P. J. & Bennett, D. (1986). The influence of genetic background and the homologous chromosome 17 on *t* haplotype transmission ratio distortion in mice. *Genetics* **114**, 235–245.

Johnson, D. R. (1974). Hairpin-tail: A case of post-reductional gene action in the mouse egg? *Genetics* **76**, 795–805.

Johnson, D. R. (1975). Further observations on the hairpin-tail (T^{hp}) mutation in the mouse. *Genetical Research* **24**, 207–213.

Klein, J., Sipos, P. & Figueroa, F. (1984). Polymorphism of *t* complex genes in European wild mice. *Genetical Research* **44**, 39–46.

Lyon, M. F. (1956). Hereditary hair loss in the tufted mutant of the house mouse. *Journal of Heredity* **47**, 101–103.

Lyon, M. F. (1984). Transmission distortion in mouse *t*-haplotypes is due to multiple distorter genes acting on a responder locus. *Cell* **37**, 629–638.

Lyon, M. F. (1986). Male sterility of the mouse *t*-complex is due to homozygosity of the distorter genes. *Cell* **44**, 357–363.

Lyon, M. F. (1987). Distorter genes of the mouse *t*-complex impair male fertility when heterozygous. *Genetical Research* **49**, 57–60.

Magnuson, T. (1983). Genetic abnormalities and early mammalian development. In *Development in Mammals* (ed. Johnson, M. H.). **5**, 209–249. Elsevier.

Mains, P. E. (1986). The *cis-trans* test shows no evidence for a functional relationship between two mouse *t* complex lethal mutations: implications for the evolution of *t* haplotypes. *Genetics* **114**, 1225–1237.

Mittwoch, U. (1989). Sex differentiation and tempo of growth: Probabilities versus switches. *Genetical Research* **53**, 227.

Phillips, S. & Nadeau, J. H. (1984). DNA preparation from mouse peripheral blood. *Mouse News Letter* **70**, 83.

Pla, M. & Condamine, H. (1984). Recombination between two mouse *t* haplotypes (t^{12tf} and $t^{Lub\text{-}1}$): mapping of the *H-2* complex relative to centromere and tufted (*tf*) locus. *Immunogenetics* **20**, 177–185.

Rigby, P. W. J., Dieckmann, M., Rhodes, C. & Berg, P. (1977). Labelling deoxyribonucleic acids to high specific activity *in vitro* by nick translation with DNA polymerase. I. *Journal of Molecular Biology* **113**, 237–251.

Schimenti, J., Cebra-Thomas, J. A., Decker, C. L., Islam, S. D., Pilder, S. H. & Silver, L. M. (1988). A candidate gene family for the mouse *t complex responder* (*Tcr*) locus responsible for haploid effects on sperm function. *Cell* **55**, 71–78.

Shin, H.-S., McCormick, P., Artzt, K. & Bennett, D. (1983). *Cis-Trans* test shows a functional relationship between non-allelic lethal mutations in the *T/t* complex. *Cell* **33**, 925–929.

Silagi, S. (1962). A genetical and embryological study of partial complementation between lethal alleles at the *T* locus of the house mouse. *Developmental Biology* **5**, 35–67.

Silver, L. M. (1985). Mouse *t* haplotypes. *Annual Review of Genetics* **19**, 179–208.

Smith, L. J. (1956). A morphological and histochemical investigation of a preimplantation lethal (t^{12}) in the house mouse. *Journal of Experimental Zoology* **132**, 51–84.

Washburn, L. L. & Eicher, E. M. (1983). Sex reversal in XY mice caused by dominant mutation on chromosome 17. *Nature* **303**, 338–340.

Washburn, L. L. & Eicher, E. M. (1989). Normal testis determination in the mouse depends on genetic interaction of a locus on chromosome *17* and the *Y* chromosome. *Genetics* **123**, 173–179.

Washburn, L. L., Lee, B. K. & Eicher, E. M. (1990). Inheritance of T-associated sex reversal in mice. (*Genetical Research*, **56**, 183–189.