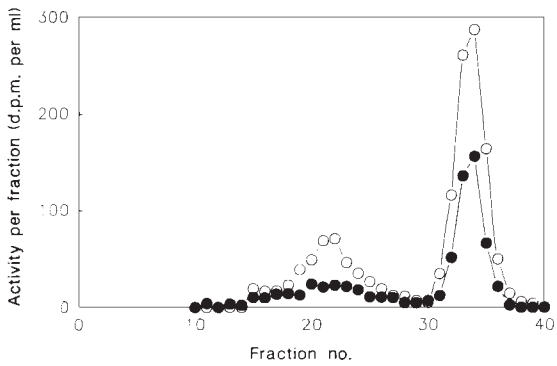

FIG. 3 Elution profiles of soluble ^{109}Cd-labelled compounds from cells grown in media containing either ^{109}Cd (activity of $Cd^{2+}=10^{-10.9}$ M) and Zn (activity of $Zn^{2+}=10^{-10.9}$ M) (●) or ^{109}Cd (activity of $Cd^{2+}=10^{-10.9}$ M) only (○). Soluble extracts were prepared as described in Table 1. Equal quantities of protein (60 μg) from each sample were applied to a Sephacryl S-200 column and the radioactivity in the eluted fractions assayed.

The Zn cell quota was the same regardless of whether or not Cd was supplied to the medium.

Three lines of evidence demonstrate that Co and Cd substitute for Zn in *T. weissflogii*. First, the growth-promoting properties of Co and Cd are only evident when Zn is absent from the medium. Second, the distribution of Co, Cd and Zn among the soluble cellular constituents is remarkably similar (leading us to believe that we are observing not an enzyme substitution but a metal substitution in the same protein). Finally, the quantity of Cd per cell decreases in the presence of Zn and increases in the absence of Zn.

Normalized to phosphorus, the cellular concentration of Cd we obtained in our experiments is in the range of those in surface seawater particles (Cd/P, $0.1\text{–}1.0\times10^{-3}$; refs 3, 22). Therefore, the biological use of Cd by phytoplankton can apparently account for its oceanographic distribution. The simultaneous depletion of several trace elements in surface sea water can be explained in part by biochemical metal substitution as exemplified here by Co, Cd and Zn. Substitution of trace metals or metalloenzymes could be a common strategy for phytoplankton survival in trace metal-improverished environments, such as the ocean, and could result in an effective colimitation of phytoplankton growth by several bioactive elements. □

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Rarity of density dependence or population regulation with lags?

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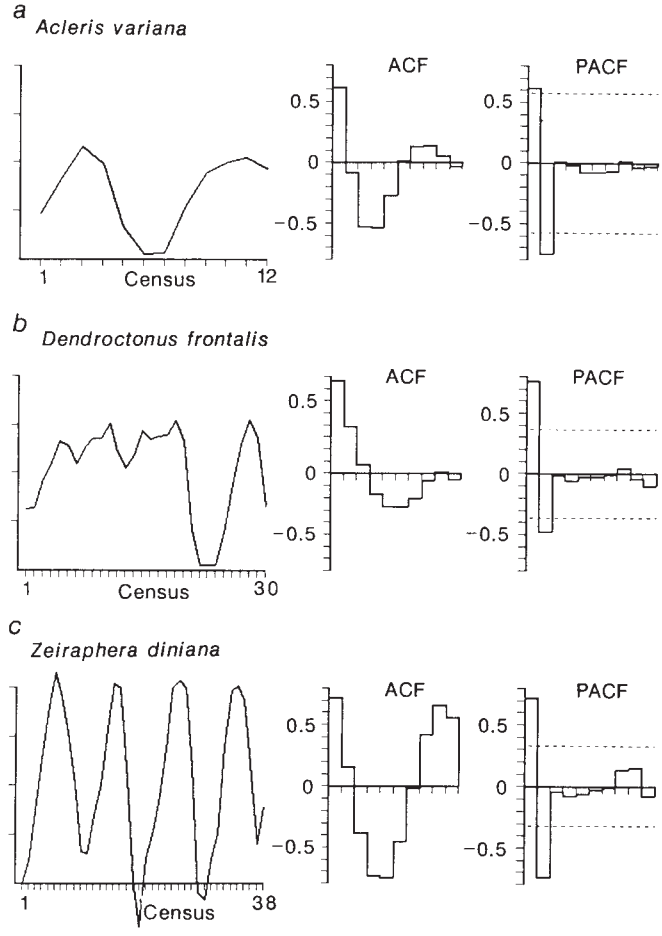
SEVERAL recent reviews of published life tables[1–3] concluded that density-dependent regulation is infrequent in insect populations, prompting a vigorous debate among ecologists[4–10]. Little attention, however, has been directed to one issue: most life-table analyses look only for direct (not-lagged) density dependence. Thus, there is a real danger that populations characterized by delays in regulation will be relegated to a density-independent limbo by an analysis not equipped to recognize such behaviour. I have evaluated the evidence for delayed density dependence in population dynamics of 14 forest insects, and assessed the effect of regulation lags on the likelihood of detecting direct density dependence. Eight cases exhibited clear evidence for delayed density dependence and lag-induced oscillations, but direct density dependence was detected in only one of these. This result suggests that traditional analyses will not, in general, detect density-dependent regulation in populations that are characterized by lags and complex dynamic behaviour.

In selecting case studies for the analysis, my focus was exclusively on temporal density dependence, which is the central issue in the population regulation controversy[5,6]. Accordingly, I looked for temporal records of forest insect populations from a single locality. When there had been a census on the same species in several locations, or there had been several studies on it, I selected the longest time-series available. I used all the records I could find of more than 10 consecutive censuses.



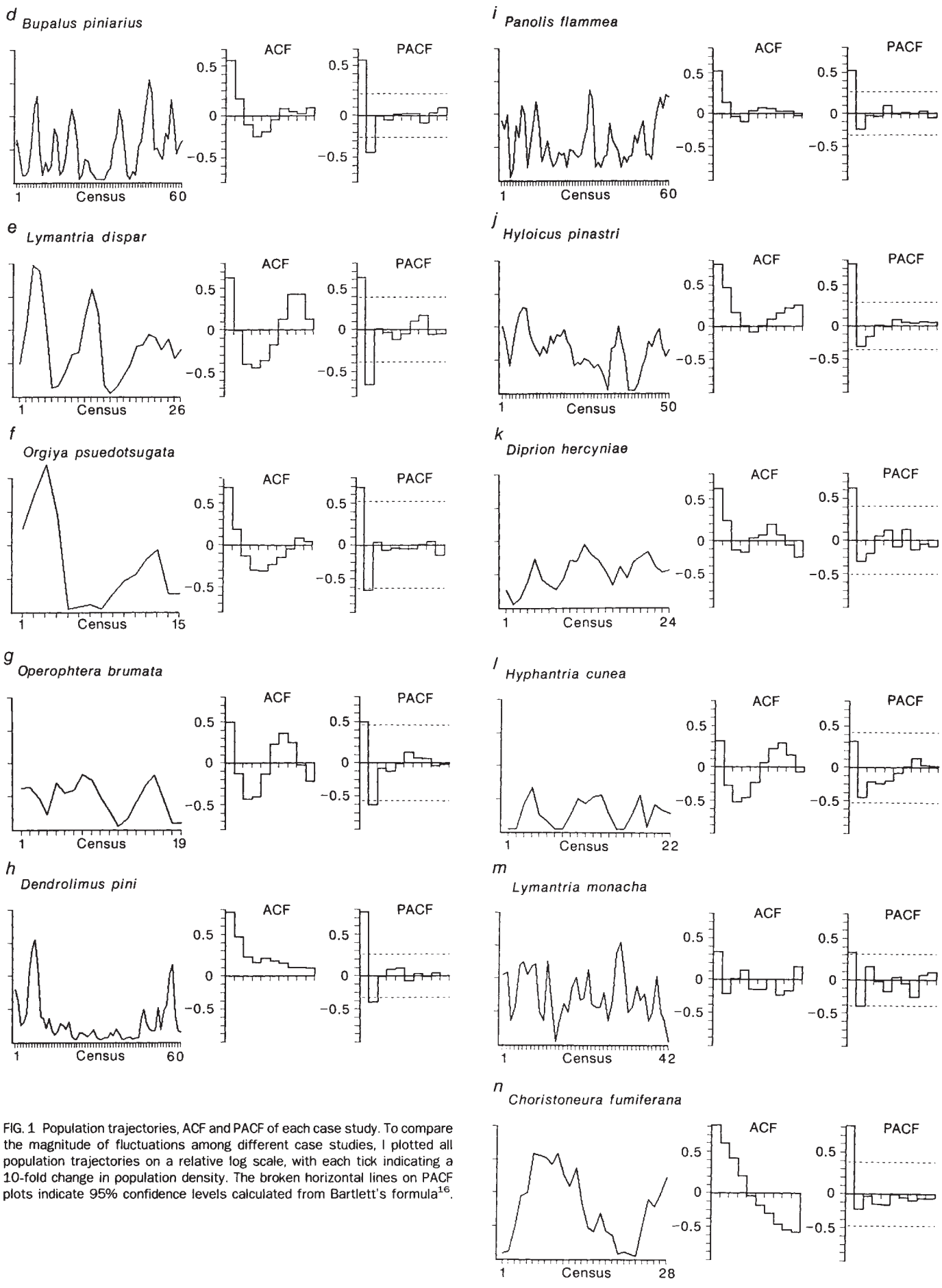


FIG. 1 Population trajectories, ACF and PACF of each case study. To compare the magnitude of fluctuations among different case studies, I plotted all population trajectories on a relative log scale, with each tick indicating a 10-fold change in population density. The broken horizontal lines on PACF plots indicate 95% confidence levels calculated from Bartlett's formula[16].

TABLE 1 Results of fitting population data with model 2

	Species	r_0	α_1	α_2	Sign	R^2
a.	*Acleris variana*[20]	0.66 (0.63)	−0.0038 (0.003)		NS	0.193
		1.26 (0.35)	−0.0015 (0.0015)	−0.0073 (0.0015)	***	0.811
b.	*Dendroctonus frontalis*†	0.66 (0.44)	−0.00028 (0.00014)		NS	0.135
		1.30 (0.35)	−0.00004 (0.00012)	−0.00055 (0.00011)	***	0.552
c.	*Zeiraphera diniana*[21]	0.54 (0.49)	−0.008 (0.004)		NS	0.087
		1.20 (0.39)	−0.0001 (0.004)	−0.020 (0.004)	**	0.511
d.	*Bupalus piniarius*[22]	0.20 (0.22)	−0.0013 (0.0005)		*	0.091
		0.34 (0.21)	−0.0005 (0.0006)	−0.0018 (0.0006)	**	0.241
e.	*Lymantria dispar*[23]	0.12 (0.44)	−0.00008 (0.00006)		NS	0.074
		0.40 (0.36)	−0.000002 (0.00006)	−0.00020 (0.00005)	***	0.426
f.	*Orgyia pseudotsugata*[24]	−0.25 (0.67)	−0.008 (0.006)		NS	0.125
		0.31 (0.40)	−0.007 (0.004)	−0.018 (0.004)	***	0.739
g.	*Operophtera brumata*[25]	0.37 (0.37)	−0.0054 (0.0030)		NS	0.183
		0.92 (0.37)	−0.0035 (0.0026)	−0.0074 (0.0027)	*	0.472
h.	*Dendrolimus pini*[26]	0.03 (0.15)	−0.0010 (0.0005)		NS	0.062
		0.10 (0.13)	+0.0002 (0.0006)	−0.0023 (0.0006)	***	0.268
i.	*Panolis flammea*[22]	0.21 (0.13)	−0.041 (0.014)		**	0.141
		0.30 (0.12)	−0.026 (0.014)	−0.040 (0.015)	*	0.232
j.	*Hyloicus pinastri*[22]	0.09 (0.09)	−0.025 (0.011)		*	0.092
		0.14 (0.09)	+0.007 (0.016)	−0.044 (0.016)	**	0.222
k.	*Diprion hercyniae*[21]	0.57 (0.24)	−0.0021 (0.0008)		*	0.252
		0.67 (0.27)	−0.0017 (0.0009)	−0.0008 (0.0009)	NS	0.286
l.	*Hyphantria cunea*[27]	0.69 (0.22)	−0.13 (0.03)		***	0.473
		0.85 (0.25)	−0.13 (0.03)	−0.04 (0.03)	NS	0.514
m.	*Lymantria monacha*[28]	0.06 (0.45)	−0.00003 (0.00002)		NS	0.055
		0.32 (0.46)	−0.00002 (0.00002)	−0.00004 (0.00002)	NS	0.125
n.	*Choristoneura fumiferana*[11]	0.29 (0.19)	−0.018 (0.009)		NS	0.139
		0.29 (0.20)	−0.016 (0.011)	−0.002 (0.012)	NS	0.140

The first line in each case study shows the results of regression on N_{t-1} only, and indicates whether this regression was significant (*, $P<0.05$; **, $P<0.01$; ***, $P<0.001$; NS, not significant). The second line shows the results of adding N_{t-2} to the regression in a step-wise fashion and testing whether the increase in regression sum of squares is significant. The numbers in parentheses are the standard errors of the estimates.

† P.T. and R. Billings, manuscript in preparation.

Delayed density dependence can arise in natural populations as a result of interactions with other members of the community, such as natural enemies[8,11], or because high population density may adversely affect the fecundity of the next generation[12]. Several methods for detecting delayed regulation have been suggested[13–15]. I used two distinct approaches, each providing a different viewpoint of the temporal population dynamics. First, I used the diagnostic techniques of time-series analysis[16]. In particular, for each record of population fluctuations I estimated the autocorrelation function (ACF) and the partial autocorrelation function (PACF). The strength of the Box–Jenkins approach lies in its ability to consider multiple lag effects. Its weakness, with respect to population modelling, is its assumption that log-transformed population densities are related in a linear fashion, for example, in a form of an autoregressive process of the order p, AR(p)

$$L_t = a_0 + a_1 L_{t-1} + \cdots + a_p L_{t-p} + \varepsilon_t \quad (1)$$

Here, $L_t = \log N_t$ is the log-transformed population density at census t; $a_1, \ldots, a_p$ are weights that quantify the influence of past densities on the population change, and ε_t is a random variable with mean zero and variance σ^2. As real-world population dynamics are inherently non-linear[11] the Box–Jenkins approach can be useful for identification of the broad patterns in the data, but not for modelling the population fluctuations.

The second approach involved specifying a non-linear model for N_t and considering a limited number of lags. I used the following phenomenological model, which is an extension of the Ricker equation,

$$N_t = N_{t-1} \exp[r_0 + \alpha_1 N_{t-1} + \alpha_2 N_{t-2} + \varepsilon_t] \quad (2)$$

The parameters of the model r_0, α_1, α_2, and σ^2 (the variance of the exogenous component ε_t) are readily estimated by regressing the rate of population change $r \equiv \log(N_t/N_{t-1})$ on N_{t-1} and N_{t-2}. This procedure assumes that errors are multiplicative and distributed log-normally, which is the standard assumption in statistical analyses of population fluctuations[11,17]. I analysed each series of population censuses using a stepwise regression: first regressing r on N_{t-1}, and then testing whether adding the term N_{t-2} significantly reduced unexplained variance.

Strong evidence for delayed density-dependent regulation was found in eight out of 14 population trajectories (*a*–*h* in Fig. 1 and Table 1). Consider the ACF/PACF graphs first. The PACF is known to be a particularly useful diagnostic tool for determining the order of an AR process[16]. The theoretical PACF of the first-order autoregressive process, AR(1), is characterized by a single spike at lag 1, and is zero everywhere else[16] (compare with the PACF shapes for cases *i* and *k*). In cases *a*–*h*, however, PACFs have two spikes (at lags 1 and 2) that are significantly different from zero (Fig. 1). Such a pattern is consistent with the theoretical PACF of AR(2) (ref. 16). Note that PACF at lag 2 is negative. A negative term involving N_{t-2} in equation (1) implies delayed density regulation. The ACF patterns in these cases also exhibited a great degree of similarity (Fig. 1). In all cases but *h*, the ACF behaved as a damped sine wave, which indicates an oscillatory endogenous component in the dynamics of populations *a*–*g*[18].

Fitting the data from these eight case studies with equation (2) showed that including N_{t-2} significantly improved the fit in all eight cases (Table 1). This result corroborates the conclusions drawn from the time-series analysis. Regression on N_{t-1} alone, however, was significant only in case *d*.

One potential problem in interpreting results of regression on r is that sampling errors in N_t bias the estimates of the slopes α_1 and α_2. Walters and Ludwig[19] showed that, for the Ricker model, the bias in the slope estimate depends on the magnitude of the true slope. If density regulation is weak or absent, then the effect of observation errors is to overestimate the amount of density dependence. On the other hand, if regulation is strong, then density dependence will be underestimated. This bias, however, does not affect my main conclusion, as it is based on a contrast between the estimated effects of direct versus delayed density dependence. In fact, observation errors make such con-

trasts conservative, as in a–h direct density dependence is weak and will therefore be overestimated, whereas delayed density dependence is strong and therefore underestimated.

For the other cases, i–n, PACF showed no evidence of delayed density-dependent regulation. But in two cases, i and j, including N_{t-2} significantly improved the fit to model (2). In cases k and l only evidence for direct density-dependence was found, whereas cases m and n showed no evidence for density-dependent regulation, either direct or delayed (Table 1).

In conclusion, an analysis that does not consider time lags will not, in general, detect density-dependent regulation in population oscillations driven by delayed density-dependent mechanisms. The analysis in this paper detected direct density dependence in only 5 out of 14 populations. This proportion, 36%, is not that different from similar proportions reported in previous surveys[1-3]. But further analysis showed that seven out of the nine apparently non-regulated populations were, in fact, subject to delayed density-dependent regulation. Thus, it is conceivable that many insect populations classified as density-independent by Stiling[3] are actually regulated by delayed density-dependent mechanisms. □

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Population structure of the human pseudoautosomal boundary

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THE mammalian sex chromosomes are composed of two genetically distinct segments: the pseudoautosomal region, where recombination occurs between the X and Y chromosomes, and the sex chromosome-specific parts[1,2]. Between these two segments the human sex chromosomes differ by the insertion of an *Alu* element on the Y chromosome[3]. We have surveyed the sequence variation in the boundary region using the polymerase chain reaction. Fifty seven Y and sixty X chromosomes from ten different human populations were analysed. The X chromosomes were found to be polymorphic at five positions in a 300-base-pair region. By contrast, all Y chromosomes were identical except for one distal polymorphism shared with the X chromosome.

We used direct sequencing and restriction mapping of DNA amplified by the polymerase chain reaction (PCR) to survey the variation between the sex chromosomes in the boundary region (Fig. 1). Position numbers were assigned by defining the *Alu* insertion site as 0 and naming the first X–Y homologous base XY1, the first Y-specific base Y1, and the first X-specific base X1. Variation at positions XY41, XY45 and XY274 has previously been described[3,4]. New variants detected in this study were found at positions X35, XY71, XY161 and XY171 (Table 1).

All the Y chromosomes were identical except for the most distal locus, XY274, where polymorphism was found in some African populations. The X chromosomes, however, showed rich genetic variation, with polymorphism detected at X35, XY71, XY161, XY171 and XY274.

At the three central loci, XY71, XY161 and XY171, two haplotypes predominated, TAT and GGA, with a third haplotype, TGA, found only in South African Bantu-speaking blacks and San (Kalahari!Kung formerly 'Bushmen'). Using data from northern Europeans, Table 2 compares the alleles at X35 and XY274 with the alleles at the three central loci as represented by XY71. Significant linkage disequilibrium was found between X35 and XY71 and between XY71 and XY274, but not between the most distant loci X35 and XY274. Table 3 summarizes the haplotype analysis of northern Europeans; of the 32 possible haplotypes, only five were found in the sample of 19 X chromosomes. More variation was found in the African San population, where five haplotypes were detected in only nine X chromosomes tested.

All Y chromosomes are different from the X chromosomes at sites XY41 and XY45. From position XY46 to XY171 the Y chromosomes have the same sequence as a common X chromosome haplotype. At position XY274 the two sex chromosomes share the same polymorphism.

The insertion of the *Alu* element must have been a unique event and all Y chromosomes in higher apes descended from the chromosome in which it happened[3]. The divergence of the X and Y chromosomes proximal to the insertion site implies that recombination has not occurred in this region since the time of the *Alu* insertion[3]. The same reduced similarity would be expected in the region distal to the *Alu* insertion if there had not been any genetic contact between the X and Y chromosomes. But it has previously been shown that the sex chromosomes are highly similar in this region[3], and in the present study we find strong evidence for recombination occurring proximal to XY274. The shared polymorphism on the X and Y chromosomes at this site would otherwise have to be explained by coincident mutations. The crossover could have occurred at any position distal to XY45, as in this region all Y chromosomes are identical with a common X chromosome haplotype. Only at positions XY41 and XY45 do we find divergence between the X and Y chromosomes. We conclude that the boundary between the sex specific and the pseudoautosomal parts of the X and Y chromosomes is at the *Alu* insertion site or very close to it.

The sequence variation found in the boundary region must be the outcome of an interplay between many evolutionary factors, including mutation, selection, recombination, population