Synaptic responses to concerted genomic evolution in *Lathyrus*. I. Intergenomic effects

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C₀ autotetraploids have been derived from seven diploid ($x=7$) species of *Lathyrus*, varying in genome size (11–20 pg DNA/2C) and content of moderately repetitive DNA (5–12 pg/2C). Pairing patterns at metaphase-I in 942 pollen mother cells (PMCs), each with a minimum of 28 chiasmata, were compared with a model relating pairing autonomy to genomic organization. Autotetraploids of one species, *L. aphaca*, form on average one multivalent more than those of the other six (5.2 vs. 3.7–4.2/PMC). With the exception of *L. aphaca*, intergenomic differences in bivalent frequency and incidence of synaptic exchange correlate with the size of the genome, particularly that of the moderately repetitive component. Such nucleotypic correlations appear to reflect losses in numbers of autonomous synaptic sites, concomitant with concerted reductions in the size of the genome. They also indicate that autonomous synapsis may be governed by moderately repetitive sequences.

Keywords: Autopolyploidy, genomic evolution, *Lathyrus*, meiosis, multivalents, synapsis.

Introduction

Eukaryotic organisms are distinguished from prokaryotes by greater complexity, larger genomes and predominantly sexual modes of reproduction. Although the size of the genome bears some relation to the level of organization in prokaryotes, it is an evolutionary maverick in eukaryotes (Cavalier-Smith, 1985). DNA is usually present in excess of the most generous estimates of genotypic requirements, often as untranscribed repetitive sequences. While variation in heterochromatic blocks of highly repetitive sequences is usually indicative of chromosomal polymorphisms (e.g. Arnold *et al.*, 1986), moderately repetitive sequences exhibit different patterns of interspersal in euchromatin which accompany phylogenetic divergence (Mizuno & Macgregor, 1974; Bouchard, 1982; Narayan, 1982). Extra DNA has variously been described as harmless junk (Ohno, 1985), parasitic (Orgel & Crick, 1980) or of nucleotypic value (Cavalier-Smith, 1985). Studies of possible nucleotypic

effects have revealed that meiosis is much more sensitive to differences in genome size than is mitosis. In flowering plants, for instance, a 25-fold increase in 2C DNA amount is accompanied by little more than a doubling of mitotic cycle time, whereas the duration of meiosis is extended by a factor of 10 (Rees & Jones, 1977, p. 33).

A crucial yet enigmatic aspect to meiosis is the ability of homologous chromosomes to associate at zygotene. In theory, a minimum of one synaptic site per chromosome would be sufficient for pairwise synapsis, giving rise to bivalents at pachytene. Although multiple pairing initiations are commonly observed in plants, the characteristics of synaptic sites only become apparent under conditions of competitive pairwise synapsis and are therefore most readily studied in autopolyploids where each chromosome has more than one potential pairing partner. In the majority of polyploids, multivalents arise through the pairwise association of chromosomes at two or more autonomous synaptic sites (Moens, 1969; Loidl, 1986; Loidl & Jones, 1986; Vincent, 1991). The autonomous nature of synaptic sites is exposed by their ability to generate changes of pairing partner in autopolyploids (Darlington, 1929). An autonomous synaptic site may be defined as the smallest region of a chromosome with the capacity to initiate changes in pairing partner in an autopolyploid (Hamey *et al.*, 1988).

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This and the following paper are dedicated to the memory of our friend and colleague J. N. Hartshorne.

If genomic evolution alters the number or spacing of autonomous synaptic sites, a mathematical model shows that such changes should have distinguishable effects on bivalent and multivalent frequency in polyploids (Callow & Gladwell, 1980; 1984). While any synaptic effects are liable to be mitigated by selection in established polyploids (Callow & Parker, 1985), they can be of little significance at the diploid level. Synaptic responses to concerted genomic evolution are therefore most likely to be detected in newly derived (C_0) autotetraploids.

Species of *Lathyrus* (Fabaceae) are ideal for nucleotypic studies of intergenomic divergence. They have a common basic number ($x=7$), are usually diploid (88 per cent) (Darlington & Wylie, 1955) but display extensive variation in genome size (7–29 pg DNA/2C) and consistent differences in genomic composition, particularly in relation to moderately repetitive sequences (Narayan, 1982; Kuriyan & Narayan, 1988). The effects of such intergenomic divergence on the mechanics of pairing autonomy have been studied by comparing the meiotic behaviour of a series of C_0 autotetraploids with the predictions of the model. The results are reported in this paper. Intragenomic comparisons are the subject of the sister paper in this series (Wallace & Callow, 1993), while influences of ploidy and chiasma variation will be explored elsewhere.

The term 'pairing partner exchange' has long been applied to both the process of exchanging pairing partners and the structural intersections to which that process gives rise (e.g. Darlington, 1929; Rasmussen, 1987). We use 'synaptic exchange' in the same sense, only modifying the phrase for simplicity and to conform with the use of 'synaptic site' and 'synaptic interval' in our chosen mathematical model (Callow & Gladwell, 1984; Hamey *et al.*, 1988). As far as structures are concerned, some authors now prefer the term 'pairing partner switch' (e.g. Loidl & Jones, 1986; Gillies *et al.*, 1987).

Model

Observations of meiotic events in C_0 autotetraploids have been tested against the predictions of a simple model which may be extended to include any type or degree of polyploidy (Callow & Gladwell, 1980; 1984). Here, we are principally concerned with inter-relations between chromosome size, numbers of autonomous synaptic sites, sizes of synaptic intervals between sites and the incidence of synaptic exchange within such intervals. Because such aspects of the model are crucial to our evidence, pertinent details of the argument are outlined below.

Assumptions

- 1 Maximal pairwise association at zygotene; any univalents and trivalents observed at metaphase-1 are thereby assumed to result from the desynaptic breakdown of bivalents or quadrivalents.
  - 2 Pairing at an average of i autonomous synaptic sites on each chromosome of a genome.
  - 3 Each synaptic interval being of similar length, θ , within a genome.
  - 4 Each synaptic interval within a genome having the same probability, ρ , of including a single synaptic exchange (Fig. 1a and b).
  - 5 The probability ρ being effectively linearly related to the length of each synaptic interval θ .
- Assumption 5 only holds for small values of θ because ρ is expected to approach the limit imposed by random pairing at large values of θ .

Derivations

- 1 The probability, α , of a set of four homologous chromosomes forming two bivalents at pachytene is the same as that of no synaptic exchange ($1-\rho$) at each of $i-1$ synaptic intervals (Fig. 1c).

Thus we have

$$\alpha = (1 - \rho)^{i-1}$$

on logarithmic transformation this yields

$$\ln \alpha = (i-1) \ln (1 - \rho). \quad (1)$$

- 2 Each chromosome has an effective synaptic length, λ , which is the sum of all $i-1$ synaptic intervals, each of length θ , giving

$$\lambda = (i-1)\theta. \quad (2)$$

- 3 As the average number of synaptic exchanges $\bar{r}$ is $\rho(i-1)$, we have

$$i-1 = \bar{r}/\rho$$

but from equation 2,

$$i-1 = \lambda/\theta.$$

We now see that the average number of synaptic exchanges is expected to be linearly related to effective synaptic length, where θ and ρ are constant because

$$\bar{r} = (\rho/\theta) \lambda. \quad (3)$$

- 4 Consistent differences in genome size may have three possible outcomes with regard to synaptic behaviour (Fig. 1d): (i) they may be unrelated to effective synaptic length, possibly contributing to chromatin loops on the periphery of synaptonemal

complexes; (ii) they may alter the number of synaptic sites i or (iii) they may change the size of synaptic intervals θ . In the first case, pachytene bivalent frequency should be unrelated to genome size. In the second, it should bear a log/linear relation to chromosome size and, in the third a log/log relation. These conclusions stem from α showing a log/linear relation to i but a log/log relation to ρ (equation 1) whereas λ is a linear function of both i (equation 2) and ρ (equation 2 and assumption 5).

Materials and methods

Seven *Lathyrus* spp. were obtained through the European Seed Exchange Scheme. C_0 autotetraploid plants

were produced by immersing germinating seedlings in a 0.01 per cent colchicine solution for 12 h. Seedlings were rinsed thoroughly under running water for 3 h before replanting in a peat-based compost. Surviving plants were grown until flowering in a glasshouse maintained at 20°C supplemented with light from sodium vapour lamps for 8 h/day. Suitable individuals for meiotic analysis were selected by examining fixed shoot-tip meristems for tetraploid cells.

Root- and shoot-tip squashes in lacto-propionic orcein were prepared after 3 h pre-treatment in 0.05 per cent colchicine, followed by fixation in ethanol/acetic acid (3:1) and staining in Feulgen. Meiotic preparations from suitably staged anthers were prepared similarly but without the colchicine pre-treatment. Thirty-six plants were examined, at least three per species (Table 1).

Estimates of pachytene pairing behaviour are based on observations of pollen mother cells (PMCs) at metaphase-I. While direct observations of other polyploids have been made using pachytene spreading (e.g. Loidl & Jones, 1986), the technique has yet to be adapted for *Lathyrus* and could not be expected to yield the large body of data required for our analysis.

Inferences of pachytene behaviour, drawn from observations of metaphase-I, are liable to be distorted by the repulsion of non-sister chromatids at diplotene, unless pairs of non-sisters are bound by chiasmata. Multivalents are particularly prone to such desynaptic disintegration (John & Henderson, 1962; Roseweir & Rees, 1962). As far as tetrasomic sets of homologous chromosomes (tetrasomes) are concerned, at least three chiasmata are required to maintain a quadrivalent to metaphase-I, whereas only two chiasmata are needed to maintain the same number of chromosomes as bivalents. Cells with seven tetrasomes therefore require at least 21 chiasmata to maintain the maximum of seven quadrivalents to metaphase-I. To increase the likelihood that each tetrasome within the cell forms at least three chiasmata, PMCs of *Lathyrus* were selected, each with a minimum of 28 chiasmata and hence a mean of at least four chiasmata per tetrasome. This should minimize lapses of pachytene pairing due to insufficient chiasmata.

Although the ensuing sample sizes varied according to the original chiasma frequency (Table 1), a large number of cells (942) was investigated with a mean very close to the selected minimum of 28 chiasmata (30.6). Studies of relations between metaphase-I pairing behaviour and chiasma frequency in the full sample of PMCs are to be published elsewhere. They indicate that the present selection of cells of high chiasma frequency does not appreciably bias the estimated range of pachytene behaviour under scrutiny. Thus, extra-

Fig. 1 Scheme of pairing at i equidistant autonomous sites in four homologous chromosomes (see Model). Two mutually exclusive pairing events are assumed to occur with probabilities of ρ and $1-\rho$ at each of the $i-1$ synaptic intervals: discordance between successive synaptic sites, producing synaptic exchange amongst all four homologues (a) and concordance, resulting in two paired segments (b). The probability, α , with which a set of four homologous chromosomes is expected to form two bivalents at pachytene is therefore $(1-\rho)^{i-1}$ because no exchange must occur (c). On logarithmic transformation, this yields $\ln \alpha = (i-1) \ln(1-\rho)$ (equation 1). Genomic increases arising from the interspersions of DNA sequences (d) may interact with effective synaptic length in three different ways: (i) by contributing only to the lateral loops of a synaptonemal complex they may make no contribution to effective synaptic length, (ii) they may increase the number of synaptic sites and (iii) they may expand synaptic intervals.

polations from the full spectrum of chiasma variation give similar estimates of pairing behaviour at high chiasma frequency to those reported here for the selected samples. Because our interest was in the presence or absence of synaptic exchange, pairs of univalents were counted as bivalents whereas single univalents accompanied by trivalents were counted as quadrivalents. Both categories were rare (Table 1).

Inferences on the incidence of synaptic exchange are especially susceptible to both chiasma frequency and chiasma distribution because each synaptic exchange will only persist to metaphase-I if individually flanked by chiasmate associations. We are therefore only able to infer minimal numbers of synaptic exchanges from our observations.

DNA amounts are those reported by Narayan (1982). Relations between bivalent frequency and DNA amount are tested in a series of regression analyses. Partial regression has been employed to detect the most critical genomic component, followed by curvilinear regression to distinguish between possibilities 4(ii) and 4(iii) in the above model.

As Narayan & Rees (1976) have pointed out, variation in the size of the genome amongst *Lathyrus* species is largely attributable to the size of the moderately repetitive component. The seven species investigated here bear testimony to this conclusion. Using the values reported by Narayan (1982), we see that moderately repetitive DNA (M) increases steeply with genome size ($1C$) ($\hat{M} = -3.513 + 0.795C$, $t_{[5]} = 12.986$, $P < 0.001$), whereas non-repetitive DNA

(N) increases only gradually ($\hat{N} = 2.708 + 0.178C$, $t_{[5]} = 3.054$, $P < 0.05$) and highly repetitive DNA (H) shows no relation to genome size ($\hat{H} = 0.805 + 0.028C$, $t_{[5]} = 0.333$, $P > 0.70$).

Results

Observations of mitotic complements confirm previous reports of variation in chromosome size reflecting amounts of total genomic DNA (Narayan, 1982). The total mitotic length in *Lathyrus hirsutus* (19.9 pg/2C), for example, shows a 36 per cent increase over that of *L. articulatus* (12.2 pg/2C) (Fig. 2) (see Wallace & Callow, 1993).

Although at least 100 cells per species were screened at metaphase-I, sample sizes of those with a minimum of 28 chiasmata varied between 29 and 391 in accordance with the degree of chiasma variation in each species (Table 1). Mean chiasma frequencies of the selected samples ranged over a very narrow band (29.0–32.4/PMC). Six of the seven species exhibited a limited spectrum of mean quadrivalent frequency (3.58–3.99/PMC). The remaining species, *L. aphaca*, formed on average one additional quadrivalent. In all cases univalents and trivalents were rare (Table 1) (Fig. 3).

The proportion (α) of tetrasomes, forming two bivalents at pachytene, is expected to attain a maximum value of one-third, under conditions of random association between homologous synaptic sites. This may be due to independent association at either end of one

Fig. 2 The diploid complements of seven species of *Lathyrus* observed at mitotic metaphase. (a) *L. angulatus*, (b) *L. articulatus*, (c) *L. aphaca*, (d) *L. cicera*, (e) *L. sativus*, (f) *L. odoratus*, (g) *L. hirsutus*. Bar = 10 μ m.

large synaptic interval or to the equivalent collective behaviour of many synaptic sites (Callow & Gladwell, 1984; Callow *et al.*, 1984). The estimated value of α in C_0 autotetraploid *L. aphaca* does not differ significantly from this prediction (0.260 2IIs/PMC, $t_{[99]} = 1.591$, $P > 0.05$). By contrast, C_0 autotetraploid *L. hirsutus*, with the next lowest estimate of α (0.405), shows an excess of bivalents and a highly significant deviation from random pairing ($t_{[390]} = 2.88$, $P < 0.01$).

With the exception of *L. aphaca*, estimates of α show a consistent decline with increasing genome size (Table 2). This decline is strictly log/linear and highly significant (Fig. 5a) (Table 3i), in accordance with the prediction of the model (4ii) where the number of synaptic sites changes with the size of the genome whereas the lengths of synaptic intervals remain constant. Partial regression analysis shows this relation to be attributable to the size of the moderately repetitive fraction (Table 3ii). Not only was the partial regression coefficient for moderately repetitive DNA the only coefficient to bear the expected negative sign but it was the only coefficient to prove significant. The logarithm

of bivalent frequency shows a highly significant linear decline with increasing amounts of moderately repetitive DNA (Fig. 5b) and gives no indication of a non-linear element (Table 3iii). These observations are consistent with the hypothesis that number of autonomous synaptic sites is sensitive to genome size (Model, 4ii) and particularly to the size of the moderately repetitive fraction.

Minimum numbers of synaptic exchanges were inferred from metaphase configurations of multivalents (Fig. 4). With the exception of a single quadrivalent in *L. aphaca* which required at least three synaptic exchanges, estimates of the minimum number of exchanges varied from one to two per multiple and from 0 to 10 per cell. The average minimum ranged from 3.73 synaptic exchanges per cell in *L. angulatus* to 5.52 in *L. aphaca*. These values represent a range of 0.532 to 0.789 *per tetrasome* (Table 2). By contrast, the minimum number of synaptic exchanges *per multivalent* remained close to unity in autotetraploids of all seven species (1.014–1.091), *L. aphaca* yielding an estimate close to the median (1.066) (Table 2).

Table 1 Average metaphase-I pairing behaviour in PMCs each with a minimum of 28 chiasmata sampled from 36 C_0 autotetraploids of seven species of *Lathyrus*

Species	Number of plants	Number of PMCs	Chiasma frequency		Mean pairing pattern			
			Mean	S.E.	I	II	III	IV
<i>L. angulatus</i>	3	62	30.3	0.31	0.26	6.56	0.10	3.58
<i>L. articulatus</i>	5	94	29.5	0.29	0.27	6.29	0.16	3.67
<i>L. aphaca</i>	8	100	29.0	0.14	0.31	3.61	0.25	4.93
<i>L. cicera</i>	4	54	29.1	0.15	0.20	6.26	0.13	3.72
<i>L. sativus</i>	4	29	29.0	0.21	0.31	5.72	0.17	3.93
<i>L. odoratus</i>	4	212	29.4	0.19	0.31	5.82	0.22	3.85
<i>L. hirsutus</i>	8	391	32.4	0.16	0.29	5.61	0.17	3.99

Fig. 3 Metaphases-I PMCs from C_0 autotetraploids of three species of *Lathyrus*: (a) *L. angulatus* ($1_1 6_{II} 1_{III} 3_{IV}$), (b) *L. aphaca* ($2_1 10_{II} 4_{IV}$) and (c) *L. hirsutus* ($4_{II} 5_{IV}$). The formation of the quadrivalents labelled 4a, 4b and 4c is depicted in Fig. 4. Bar = 10 μ m.

The incidence of minimal synaptic exchange increased significantly with the size of the genome, again excluding *L. aphaca* (Fig. 6a) (Tables 2, 3iv). Although none of the partial regression coefficients for the various DNA components proved significant, that

for moderately repetitive DNA came closest and was the only coefficient to bear the appropriate positive sign (Table 3v). Indeed, a separate regression against the size of the moderately repetitive component by itself proved significant (Table 3vi) (Fig. 6b). This final

Table 2 Proportions (α) of tetrasomes forming two bivalents at pachytene and average minimal numbers of synaptic exchanges ($\bar{r}$) per tetrasome, estimated from C_0 autotetraploid *Lathyrus* PMCs each forming at least 28 chiasmata and compared with estimates of genomic composition

Species	DNA composition* pg/unreplicated chromosome				Bivalent frequency		Synaptic exchange		$\bar{r}/\text{mult.}$
	Non	Mod.	High	Total	α	s_α	$\bar{r}$	s_r	
<i>L. angulatus</i>	0.31	0.35	0.11	0.77	0.475	0.063	0.532	0.065	1.014
<i>L. articulatus</i>	0.37	0.47	0.03	0.87	0.453	0.051	0.570	0.056	1.042
<i>L. aphaca</i>	0.36	0.50	0.14	1.00	0.260	0.044	0.789	0.052	1.066
<i>L. cicera</i>	0.41	0.55	0.04	1.00	0.450	0.068	0.601	0.079	1.091
<i>L. sativus</i>	0.36	0.74	0.10	1.20	0.414	0.091	0.635	0.107	1.083
<i>L. odoratus</i>	0.41	0.70	0.10	1.21	0.419	0.034	0.628	0.039	1.079
<i>L. hirsutus</i>	0.46	0.87	0.09	1.42	0.405	0.025	0.612	0.026	1.029

*After Narayan (1982).

Table 3 Multiple regression analyses of relations between average meiotic pairing at high chiasma frequency and genomic composition in C_0 autotetraploids representing six species of *Lathyrus*: *L. angulatus*, *L. articulatus*, *L. cicera*, *L. odoratus*, *L. sativus* and *L. hirsutus*. These regressions test predictions of the pairing model (see Fig. 1). DNA amounts are in pg/unreplicated chromosome (see Table 2)

- (i) Effect of total DNA on bivalent frequency
 $\ln \alpha = -0.561 - 0.251 \text{ total DNA}; \quad t_{[4]} = -8.463; \quad P < 0.01$
 (deviation from linearity, $F_{[1/3]} = 0.974, P > 0.05$)
- (ii) Effects of DNA fractions on bivalent frequency
 $\ln \alpha = -0.701 + 0.242 \text{ non rep.} - 0.376 \text{ mod. rep.} + 0.080 \text{ high rep.}$
 $t_{[2]} = 1.070 \quad t_{[2]} = -6.059 \quad t_{[2]} = 0.363$
 $P > 0.30 \quad P < 0.05 \quad P > 0.70$
- (iii) Effect of moderately repetitive DNA on bivalent frequency
 $\ln \alpha = -0.635 - 0.321 \text{ mod. rep. DNA}; \quad t_{[4]} = -12.267; \quad P < 0.01$
 (deviation from linearity, $F_{[1/3]} = 0.907, P > 0.05$)
- (iv) Effect of total DNA on minimal synaptic exchange
 $\bar{r} = 0.452 + 0.134 \text{ total DNA}; \quad t_{[4]} = 2.974; \quad P < 0.05$
- (v) Effects of DNA fractions on minimal synaptic exchange
 $\bar{r} = 0.585 - 0.307 \text{ non. rep.} + 0.255 \text{ mod. rep.} - 0.366 \text{ high rep.}$
 $t_{[2]} = -0.586 \quad t_{[2]} = 1.788 \quad t_{[2]} = -0.656$
 $P > 0.30 \quad P > 0.10 \quad P > 0.30$
- (vi) Effect of moderately repetitive DNA on minimal synaptic exchange
 $\bar{r} = 0.489 + 0.175 \text{ mod. rep. DNA}; \quad t_{[4]} = 3.366; \quad P < 0.05$

Fig. 4 Pachytene multiples inferred from the metaphase configurations of three quadrivalents in C_0 autotetraploid *L. hirsutus* (see Fig. 3c). These quadrivalents give evidence of (a) a single non-localized synaptic exchange, (b) a single synaptic exchange localized between two distal chiasmata and (c) two synaptic exchanges within the same chromosome arm. Bar = 5 μ m. Pachytene configurations are not drawn to scale.

relation concords with equation 3 and implies that amounts of moderately repetitive DNA are correlated with differences in effective synaptic length. Estimates of minimal numbers of synaptic exchanges and any analyses on which such estimates are based remain of limited value. A direct study based on pachytene spreads could provide sufficient data for measurement of the relation between the total incidence of synaptic exchange and the size of the genome. Such evidence

would provide a useful test of the validity of assumption 5.

Discussion

Chromosomal synapsis must represent the culmination of a complex hierarchy of molecular and structural interactions. At the molecular level, autonomous synaptic sites may be represented by interspersed specialized sequences consisting partly of conserved functional repeats and partly of unique regions of high pairing specificity. Alternatively, any random segment of genomic DNA may have the potential to pair autonomously. The possibility of specialized sequences is supported by the present paper, which implicates a moderately repetitive fraction in pairing autonomy. The nature of such sequences remains unknown. Some indication may be afforded by the findings of Stern & Hotta (1987) but corroborative evidence is lacking. Observations of post-meiotic segregation and gene conversion support the concept of intimate pairing between non-sister coding sequences, leading to the formation of heteroduplexes (Hastings, 1987). This may follow the association of any autonomous synaptic sites and may even take place between chromatin loops outside the synaptonemal complex.

At the structural level, only up to about 0.3 per cent of the genome can be accounted for in the lateral elements of synaptonemal complexes (Lie & Laane, 1982; Jones *et al.*, 1983; John, 1990). Most of the chromatin is present as a series of uncomplexed loops (von Wettstein, *et al.*, 1984). Synapsis in plant meiocytes begins at numerous independent chromosomal locations and the formation of synaptonemal complex is mediated by distinct pairing nodules (Jones & Albini, 1988).

The autonomous nature of synaptic sites is revealed by synaptic exchange in polyploids (Darlington, 1929; Hamey *et al.*, 1988). The reported incidence of synaptic exchange in triploid *Allium sphaerocephalon* (Loidl & Jones, 1986) and tetraploid *A. vineale* (Loidl, 1986) has been shown to follow a Poisson frequency distribution (Hamey *et al.*, 1988). A similar analysis has confirmed that synaptic exchanges show Poisson frequency distributions in autopolyploid *Crepis capillaris* (J. E. Vincent & G. H. Jones, personal communication). In all these cases, therefore, we have evidence of a rare event being equally likely at a large number of stations. The implication of such evidence is of large numbers of synaptic intervals, each with a low probability of synaptic exchange (Hamey *et al.*, 1988). Under such conditions, synapsis will be quasi-continuous (Callow & Gladwell, 1984; Hamey *et al.*, 1988).

Concerted reductions in genome size during the evolution of *Lathyrus* have been accompanied by loss

Fig. 5 Relations between the natural logarithm of the bivalent frequency ($\ln \alpha$), (a) total genomic DNA content and (b) moderately repetitive DNA content in seven *Lathyrus* species (see Table 3). Samples are based on PMCs with a minimum number of 28 chiasmata. Regression analysis does not include *L. aphaca*. (A) *L. angulatus*, (B) *L. articulatus*, (C) *L. aphaca*, (D) *L. cicera*, (E) *L. sativus*, (F) *L. odoratus*, (G) *L. hirsutus*.

of some non-repetitive DNA but have been achieved largely at the expense of moderately repetitive material (Narayan, 1982; Kuriyan & Narayan, 1988). Six of the seven species investigated here constitute a nucleotypic series, within which the number of autonomous synaptic sites declines with genome size but the spacing of such sites remains constant. The extent to which the members of this series represent an evolutionary progression, remains unknown.

Three features of our analyses point to the moderately repetitive genomic component as the source of autonomous synaptic sites. Firstly, although non-repetitive and moderately repetitive DNA are both significant correlates of genomic size in *Lathyrus*

(Narayan, 1982), only the latter genomic component yields a significant decline in bivalent frequency. The partial regression coefficient relating bivalent frequency to the size of the non-repetitive component is not only non-significant but positive. Secondly, the six species of the nucleotypic series give identical rankings for bivalent frequency and the amount of moderately repetitive DNA but not for genome size. Thirdly, only the size of the moderately repetitive genomic component yields the predicted positive relation with the average minimum number of synaptic exchanges.

Should autonomous synaptic sites reside in the moderately repetitive component, they could be interspersed throughout the genome and yet include core

Fig. 6 Relations between the average incidence of synaptic exchange ($\bar{x}$), (a) total genomic DNA content and (b) moderately repetitive DNA content in seven *Lathyrus* species (see Table 3). Samples are based on PMCs with a minimum number of 28 chiasmata. Regression analysis does not include *L. aphaca*. Species codes are as in Fig. 5.

sequences of common function. Synaptic sequences may or may not be transcribed because pairing initiation could conceivably occur without transcription.

Although the taxonomy of *Lathyrus* species is in need of review, such treatments that exist all place *L. aphaca* in a distinct subsection of the genus (Senn, 1938; Davis, 1970). Our finding that this species has a distinctive pattern of meiotic behaviour therefore accords with the possibility of a separate phyletic origin. Indeed, a genus as large as *Lathyrus* is very likely to be represented by several nucleotypic series of the type reported here. While we can as yet have no inkling of the mechanical nature of any differences between the nucleotypic series, some pointers are

available from our data. Despite their variation in quadrivalent frequency, C_0 autotetraploids of all seven species gave very similar estimates of the minimum number of synaptic exchanges per multivalent. Even *Lathyrus aphaca* with its distinctively high quadrivalent frequency showed only an average minimum of one synaptic exchange per multiple. Apparently, therefore, the genome of *L. aphaca* is neither especially rich in autonomous synaptic sites nor does it appear likely to contain unusually large synaptic intervals. The evidence of random association in this species may, however, point to a high level of homozygosity for pairing sequences (Callow *et al.*, 1984).

Previous intergenomic comparisons of *Lathyrus* species have revealed no evidence of a nucleotypic relation between chiasma frequency and genome size (Rees & Durrant, 1986). Such comparisons are, however, liable to be confounded by influences of selection at the diploid level, particularly as reductions in genome size have been accompanied by transitions from outbreeding to inbreeding (Rees & Dale, 1974). Changes in pairing autonomy are, by contrast, unlikely to be affected by selection, being detected in newly synthesized autotetraploids and of no adaptive significance to the original diploids.

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