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Dosage compensation and sex-chromatin in non-mammals

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There is an imposing body of evidence (McKusick, 1962; Davidson, Nitowsky & Childs, 1963; Lyon, 1963; Russell, 1963) to support the view that in the somatic cells of female mammals one or other X chromosome is genetically inactivated. Determination of which X is inactivated in a particular cell is believed to occur at random in early embryonic life, and all descendants of that cell maintain inactivation of the same X, thus resulting in mosaic expression of autonomous sex-linked genes in heterozygous females. This has been shown for mice, cats and humans, and may well apply to all mammals. In view of the results of Russell (1963) on mice, and of the abnormality of Turner's syndrome (XO) in humans, the simple hypothesis probably needs to be qualified to cover inactivation of part, rather than of the entire X. It is also widely accepted that the sex-chromatin or 'Barr body' of somatic interphase nuclei, the 'drumstick' of polymorph leucotyces, and the heteropyknotic (Ohno, Kaplan & Kinosita, 1959) and 'hot' (Rowley *et al.*, 1963) Xs of prophase nuclei all represent the genetically inactivated X.

This inactivation of an X has often been described as 'dosage compensation', a term originally introduced by Muller (1932) in relation to *Drosophila*. While X-inactivation is a mechanism of dosage compensation in the sense that it equalizes the effective dosage of sex-linked genes in the two sexes, it has sometimes been overlooked (e.g. McKusick, 1962; Davidson *et al.*, 1963) that the mechanism of dosage compensation in *Drosophila* is radically different from the X-inactivation of mammals. Sex-linked genes in *Drosophila* do not normally show mosaic expression in the heterozygote, and the variegated (mosaic) effects associated with translocations (Lewis, 1950) serve only to reinforce the fact that in normal females both Xs are active together. Nevertheless, in most (not all) sex-linked mutants in *Drosophila* the hemizygous male phenotypically resembles the homozygous female; dosage compensation apparently extends to sex-linked polygenes affecting the number of sternopleural chaetae (A. Robertson, personal communication, 1963), and the results of Robertson & Reeve (1953) indicate, on the whole, that the same is true for wing length. Stern (1929, 1960) and Muller (1932, 1950) have postulated the existence of special X-borne 'compensatory modifiers' to account for this. These modifiers (in general, a separate set of modifiers for each compensated locus) act to reduce the effects of the compensated gene, and the extra dose of modifiers in the homogametic sex cancels out the extra dose of the compensated

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gene. Irrespective of the exact details of compensation in *Drosophila*, a mechanism of this general type, which acts on gene products, rather than on genes or whole chromosomes, and which is effective at a cellular level, may be termed dosage compensation *sensu strictu*. This definition excludes the X-inactivation of mammals, which achieves dosage compensation (in the broad sense) at the level of the individual, but avoids the need for it at the cellular level, since only one allele is active in any one cell.

Although the basic ideas of the theory of dosage compensation were first put forward by Stern, it will be convenient here, in view of Muller's subsequent elaboration and experimental testing of the theory, to refer to it as Muller's theory. Muller argues that the compensatory modifiers have accumulated under the influence of natural selection; many sex-linked genes affect characters which have no other connexion with sex, and the optimum effective dosage of these genes will be the same, or nearly the same, in the two sexes. It is compensation of the effects of wild-type genes which is selectively important; compensation of comparatively rare mutants is a conspicuous but selectively irrelevant by-product. The mechanism whereby compensation is achieved is also irrelevant to selection; to this extent, the discovery of X-inactivation in mammals is a confirmation of Muller's selective argument. Goldschmidt (1938, 1954, 1955), on the other hand, has argued that there is no need to postulate a specially selected system of modifiers. The two sexes represent different developmental systems, and the same dose of a gene will not, in general, produce the same effect in the two systems; dosage compensation, when it occurs, is an automatic result of the different developmental systems. While it is to be expected that different reactivities of this kind will sometimes occur, it appears as a strange coincidence that their direction and magnitude should (in *Drosophila*, at least) so often be such as to cancel out exactly the effects of the difference in gene dosage. For more specific obstacles to accepting Goldschmidt's views as a general explanation of dosage compensation in *Drosophila* see Muller (1950) and Stern (1960).

The question of whether dosage compensation (either *sensu strictu* or X-inactivation) occurs in groups other than mammals and *Drosophila* has been relatively neglected. The facts discussed below are not new, nor does their collation lead to any radical resolution of difficulties, but it will perhaps serve to emphasize that the analysis of dosage compensation is 'not yet a closed chapter' (Stern, 1960), and to encourage workers on diverse groups of animals to look for other relevant facts.

1. GENETICAL EVIDENCE IN BIRDS AND LEPIDOPTERA

In organisms which lack the facilities of *Drosophila* for adding or subtracting extra doses of a gene in chromosome fragments, it is less easy to decide whether dosage compensation *sensu strictu* occurs. While sexual inequality of expression of a sex-linked gene is (provided the wild-types of the two sexes are equal in the character affected) *prima facie* evidence that compensation is absent (or incomplete), it is more difficult to prove that compensation does occur in a particular instance (see Cock, 1953, Table 2). For example, sexual equality in a recessive *may* be due to

dosage compensation, but it may be due to the mutant being an amorph; this can be excluded only if there are multiple alleles with quantitatively graded effects. Two-allele recessives of equal expression and 'recessive' lethals where the homozygous type is unknown are therefore omitted from the examples given below. Partially sex-linked genes, e.g. in fishes (Gordon, 1957), are also omitted, since, as in the classical case of *bobbed* in *Drosophila* (Stern, 1929) there is no need for dosage compensation when an allele is present on the Y chromosome.

In domestic fowls, females hemizygous for the sex-linked semidominant *B* (barred plumage) phenotypically resemble heterozygous males; homozygous males are much more extreme. This pattern of expression is characteristic of an uncompensated neomorph (i.e. a gene whose wild-type allele has no effect on expression of the mutant), and other evidence (Cock, 1953) supports the view that *B* is a neomorph. The same pattern of expression is shown by *Sd* (sex-linked dilution; probably a more extreme allele of *B*) (Munro, 1946; Albada & Kuit, 1960) and by *K* (slow feathering) (Siegel, Mueller & Craig, 1957). *B* and *K* are autonomously expressed in skin grafts, independently of the sex of the host (Danforth, 1929, 1939), although it has not been proved that this autonomy extends to the *K*/*K* versus *K* or *K*/+ difference. The results of Cock & Morton (1963) on a sex-linked effect on size and conformation favour lack of dosage compensation of the gene(s) involved, although they are not in this respect conclusive. There is a mosaic splashing effect associated with *B* (Serebrovsky, 1925; Cock, 1953) which, although superficially similar to that occurring in mice with X-autosome translocations (Russell, 1963), shows conclusively that both Xs are active in the great majority of cells. Serebrovsky's work, incidentally, antedates by 38 years—and betters by nearly 50 recombination units—Lyon's (1963) claim that her findings with *tabby* and *striated* in mice 'must be the first time in genetics that cis and trans heterozygotes for two completely non-allelic genes, ten units of recombination apart, have been shown to be phenotypically different'.

In pigeons B^{St} (almond) and B^{Of} (faded) are also expressed as uncompensated neomorphs (Hollander, 1942); both homozygotes are practically white, whereas the heterozygotes (B^{St}/+ and B^{Of}/+) are phenotypically similar to the corresponding hemizygotes. A mosaic splashing effect (Hollander & Cole, 1940; Hollander 1944) again confirms that both Xs are active in the great majority of cells. The recessive *cinnamon* in budgerigars (Taylor & Warner, 1961) is expressed as an uncompensated hypomorph (the mutant has an effect similar to, but quantitatively less than, that of the wild-type allele). Black melanin is replaced by brown in *cinnamon* birds; two doses in the male give a distinctly darker shade of brown—a closer approach to the wild-type black—than one in females. In canaries, the otherwise similar *cinnamon* mutant apparently has no marked sexual difference in expression (Gill, undated). The Pilgrim breed of geese, unlike most other domestic breeds of geese, is sexually dimorphic in plumage colour: females are pale grey, males white. Jerome (1959) has attributed this to a sex-linked dilution gene: *Sd* or *Sd*/+ gives pale grey, *Sd*/*Sd* gives white. His results from F_1 breed crosses give some support to this, but it cannot be regarded as conclusively proved (see

Lühmann, 1954). Sex-linked slow-feathering occurs in turkeys (Asmundson & Abbott, 1961), but test-matings to discover whether (as with the similar gene in fowls) K/K is more extreme than K and $K/+$ have not been made.

Three avian sex-linked loci with multiple alleles showing no sexual difference in expression have been reported: $+^S$ (gold), S (silver) and S^{al} (semialbino) in fowls (Werret, Candy, King & Sheppard, 1959; Cole & Jeffers, 1963); $+^n$ (bronze), n (narragansett, i.e. pale bronze) and n^{al} (semialbino) in turkeys (Asmundson, 1950) and $+^c$ (black), c (chocolate) and c^A (ash-red) in pigeons (Hollander, 1944). Except in the case of S, where there is also mutational evidence, the assignment of the three alleles to one locus rests on failure to detect crossing-over. If these are really single loci, the fact that males homozygous for the phenotypically intermediate alleles (S, n and c) are identical (allowing for hormonally controlled sexual differences in pattern, also present in the wild-types) with the corresponding hemizygotes might be thought to show that dosage compensation occurs. However, the dominance relations—$S > +^S > S^{al}$ and $c^A > +^c > c$, with the phenotypically intermediate allele at one end of the dominance series—clearly exclude any simple dosage model, with or without compensation; the effects of the alleles at each locus do not differ solely in a single quantitative respect (cf. Stern, 1943). In turkeys, $+^n > n > n^{al}$; the fact that n/n is phenotypically similar to n/n^{al} as well as to n (Asmundson, personal communication, 1964) may indicate merely that changes in dosage over this range are too small to have clearly distinguishable effects. None of these loci can therefore be regarded as giving evidence either for or against dosage compensation. The examples given earlier suggest that absence of dosage compensation may be widespread or even universal in birds, although they are insufficiently numerous to establish this firmly.

A search of the literature on other animal groups has revealed only two critical examples, both of them in Lepidoptera. Goldschmidt's (1921) semidominant gene (C) for melanism in *Lymantria monacha* is expressed as an uncompensated neomorph. (This may not be a very good example, since the autosomal melanic gene B is more strongly expressed in males, suggesting that males may be more sensitive to melanization generally. If this is so, some degree of dosage compensation might be required to explain why $C/+$ males are not even more extreme than C females.) Stehr's (1959) scheme for explaining the inheritance of haemolymph colours in *Choristoneura* spp. involves a balance between alleles of differing strengths at two antagonistic loci; one autosomal, the other sex-linked. Stehr tacitly assumes that there is no dosage compensation at the sex-linked locus, and this assumption appears to be an indispensable part of any hypothesis capable of explaining his extensive breeding results. Stehr also points out that his scheme, by changes in allelic strength, or fixation of one or other locus, can lead, *inter alia*, to various forms of sex-limited inheritance, and suggests that this mechanism underlies many or all of the sex-limited polymorphisms which are so common in Lepidoptera. If true, this would imply that absence of dosage compensation is widespread in Lepidoptera, but Sheppard (1961) has pointed out difficulties in accepting this part of Stehr's argument.

No examples have been found (outside *Drosophila* and mammals) of genes which *certainly* are dosage compensated. This could reflect merely the relative scarcity of known sex-linked genes in other groups, and the greater difficulty of proving than of disproving dosage compensation in a particular instance. However, the examples given above at least show that dosage compensation is not so universal a phenomenon as has sometimes been supposed. It is difficult to discern any particular significance in the fact that all the examples of absence of dosage compensation come from species in which the female is the homogametic sex. It may be relevant that most of the examples (all except *cinnamon* in canaries and haemolymph colours in *Choristoneura*) are mutants of neomorphic expression. Muller's selective argument does not apply to such genes, at least not in its simplest form; nevertheless some neomorphic mutants in *Drosophila*, notably *Bar*, are compensated. Absence of dosage compensation might be explained on Muller's theory by a special selective situation; thus the selective advantages of sex-limited polymorphism in some Lepidoptera (Sheppard, 1961) might involve the abolition or reversal of the usual selective advantages of dosage compensation. Muller (personal communication, 1963) suggests that absence of dosage compensation in Lepidoptera and birds is due to the X-chromosome having only recently been transformed from an autosome, so that there has not yet been time for selection to accumulate compensatory modifiers. A similar argument has been used (Muller, 1950) for the right arm of the X of *Drosophila pseudoobscura*, but the evolutionary time-scale involved seems far too long for this to be plausible for birds or Lepidoptera.

2. EVIDENCE FROM SEX-CHROMATIN

Ohno, Kaplan & Kinosita (1960), Ishizaki & Kosin (1960) and Moore & Hay (1961) found sex-chromatin in various tissues of female fowls, but not in males. Ohno *et al.* also found that the single X of the female is (like one of the Xs of the female mammal) positively heteropyknotic in early prophase. Beckert (1962) reports that typical sex-chromatin was found exclusively in females in several species of birds and two reptilian species, but not in Amphibia. In leucocytes, however, 'drumsticks' were found to be characteristic of the *homo*gametic sex in birds, reptiles and Amphibia. Beckert therefore believes that, contrary to the mammalian evidence, drumsticks and sex-chromatin are not homologous structures; drumsticks are associated with the homogametic sex; sex-chromatin with the female. (Beckert does not state the actual species studied; the identity of the homogametic sex is uncertain in reptiles and variable in Amphibia (van Brink, 1959).) Other investigators have failed to find any sexual dimorphism in somatic nuclei of fowls (Biggs & Payne, 1961; Miles & Strong, 1962) or of other birds and reptiles (Ashley & Theiss, 1959). Schmid (1962) found that the X in fowls did not differ in its DNA replication pattern from the major autosomes, i.e., there was no 'hot' X in either sex. The conflict of evidence as to the occurrence and nature of sex-chromatin in non-mammalian vertebrates may be due to differences in cytological technique, or, in the case of Biggs & Payne, to the peculiar difficulties of the

material used; it can only be resolved by further work. However, the genetical evidence suggests that sex-chromatin representing an inactivated X should occur in neither sex in birds; not in males, since there is no dosage compensation, and certainly not in females, since this would imply the complete suppression of expression in females of all or most sex-linked genes. Schmid's observations give some support to the possibility that the sex-chromatin represents a Y chromosome.

In Lepidoptera, Smith (1945) has shown that a very clearly marked chromatin mass occurs in the interphase nuclei of several somatic tissues of females of *Choristoneura* (*Archips*) *fumiferana*. Cytologically, this seems remarkably similar to the sex-chromatin of mammals, but again the genetical evidence (Stehr, 1959) is against the possibility that it represents an inactivated X. A similar nuclear body occurs in females of *Bombyx mori* (Jucci, 1948), *Peronia variana*, *Datana ministra* and *Hyphantria textor*, but is absent in *Malacosoma disstria*, *M. pluviale*, *M. americana* and *Rhyacionia buoliana* (S. G. Smith, personal communication, 1963); the possibility that it occurs only in species with a Y chromosome has not been tested. In many Heteroptera (Geitler, 1939), the Y chromosome is heteropyknotic in somatic cells of the male, and is visible in interphase nuclei as a sex-chromatin-like body (or bodies; somatic polyploidy is common). In other species, with XO males, there is no 'sex-chromatin' in either sex, but in *Gerris lateralis* (XO males) the exceptionally large X is similarly heteropyknotic, so that female cells have twice as many sex-chromatin bodies as male cells of the same ploidy.

3. MECHANISMS OF DOSAGE COMPENSATION

The mechanism of dosage compensation *sensu strictu* in *Drosophila*, as opposed to the fact of its occurrence, is in some respects obscure. In the salivary glands the single X in the male is approximately twice as thick as each of the Xs in the female or the autosomes of either sex, (Dobzhansky, 1957), although its DNA content is not increased. Dobzhansky suggests that this enlargement of the X is due to the accumulation of some products of gene activity, and is the cytological counterpart of dosage compensation; the single X in the male 'works twice as hard as does each of the two Xs in the female'. It would be unwise to assume that what is true of the salivary glands is necessarily true of other tissues. In any case, Muller's theory implies that dosage compensation operates by reducing the effective action of sex-linked genes in the female, not by enhancing their action in the male. This difficulty could be avoided by assuming that it is the *absence* of X-enlargement in the female which represents the compensation phenomenon, but the theory also requires that the X chromosome should contain three types of gene, each distributed more or less at random along the X: (i) the sex-determining genes, (ii) the 'ordinary' sex-linked genes, and (iii) the compensatory modifiers. Of these, only (ii) can be compensated, whereas enlargement of the X in the male salivary gland (or its absence in the female) is something which affects the whole X uniformly. (Not all bands in the X are equally enlarged, but the 'puffy' phenomenon follows the same pattern in physiologically similar cells of male and female: Rudkin, in discussion of Beerman (1956). This is superimposed on the general enlargement of the X in

males.) Since each locus of type (ii) is supposed, in general, to have several compensators, it must presumably often happen that a gene of type (ii) functions also as a compensator for another type (ii) locus; such genes must be compensated in their 'main' effects, but not in their compensatory effects. Complicated as this may seem, it is not impossible, as the modifying effect of w^+ (and *some* other *w*-alleles) on the near-by *zeste* locus (Gans, 1952) shows. In its effects as a modifier of *zeste* (actually a 'decompensating', not a compensating modifier) w^+ is not dosage-compensated; in its 'main' effect on eye colour, it is compensated. A somewhat analogous complication occurs with the mutants in *Hairy-wing* (see Cock, 1953) and *scute* (Lieb, 1942), where the degree of dosage compensation can vary independently in different regions of the body.

There seems, however, to be no compelling reason to assume, as Muller does, that *all* the compensatory modifiers must themselves be carried on the X. It is admittedly less easy to conceive a simple model of how an autosomal compensator could operate, but autosomal compensators do appear to exist. Thus the 4th chromosome modifier *m*(*B*)4 of the sex-linked *Bar* (Bridges & Brehme, 1944) increases eye size in hemizygous *Bar* males to nearly that of the normal heterozygous female, but females (homozygous and heterozygous) are unaffected, so that, in the presence of *m*(*B*)4, *Bar* is dosage compensated only slightly, if at all. The normal allele of *m*(*B*)4 is therefore a compensatory modifier of *Bar*. (The possibility that the results of Lieb (1942; see Muller, 1950) on the effects of various 1–4 translocations on the expression of *Bar* and of other sex-linked genes may be partly due to differences in dosage of the 4th chromosome, rather than of the X, thus appears to be a very real one.) The 3rd chromosome mutant *m*(*B*)3 (Steinberg, 1941) similarly, but less markedly, reduces the degree of dosage compensation of *Bar*, although not of the *Double-Bar* and *Infra-bar* 'alleles' of *Bar*. Those more familiar with *Drosophila* could probably provide further examples. Autosomal compensators presumably operate by interacting with another sex-linked locus or loci, and it could be argued that these sex-linked loci are the 'real' dosage compensators. But from an evolutionary point of view, it remains true that the whole genome, and not merely the X chromosome, is exposed to selection favouring the development of dosage compensation. Since selection acts on second-order effects only, this does increase the plausibility of the selective argument. Mammals have presumably evolved at some stage from animals in which there was no inactivation of an X in females; the genes which brought about this change are, in a sense, dosage compensators, but there is no reason to suppose that they are exclusively sex-linked. The admission of autosomal as well as sex-linked dosage compensators brings one nearer to Goldschmidt's views, although it does not involve either his minimizing of the role of selection, or his identification of the compensation mechanism with sex itself.

4. DOSAGE COMPENSATION IN SPECIES WITH IMPATERNATE MALES

Males in these species are of haploid origin; females are biparental and thus of diploid origin. ('Haploid' and 'diploid' will be used with inverted commas to

denote the origin of the individual; without inverted commas in referring to the actual chromosome content of particular tissues.) If this situation is retained in somatic tissues, the dosage of all genes will be twice as great in one sex as in the other, as with sex-linked genes in other organisms. Stern (1960) has suggested that somatic diploidy or polyploidy in the male is a mechanism of dosage compensation in some of these species, specifically *Habrobracon* and the honey bee (*Apis*). Two questions arise in this connexion. How far are somatic tissues in the male in fact diploid or, more generally, of the same degree of ploidy as corresponding female tissues? How far does the haploid–diploid situation require any special mechanism of dosage compensation?

In *Habrobracon*, at least, genetical evidence indicates that 'haploid' males are indeed somatically haploid (or of half the degree or ploidy of females). The recessive mutant *fused* produces fusion of the segments and shortening of the antennae and tarsi, and shortening of the wings. It is much more weakly expressed in 'haploid' males than in females, but biparental 'diploid' *fused* males (homozygous at the sex-determining 'locus') are similar to *fused* females (Whiting, 1943*a*). It is difficult to see an alternative to Whiting's conclusion that these differences are due to the different dosage of *fused*. The expression of most other mutants in *Habrobracon* (Whiting, 1932) and in *Apis* (Drescher & Rothenbuhler, 1963) appears to be equal, or nearly equal in females and 'haploid' males; the interpretation of this would depend on the expression in 'diploid' males, but this has apparently not been tested. The usual techniques for producing and detecting 'diploid' males in *Habrobracon* (Whiting, 1943*b*), or mosaics with 'diploid' male parts in *Habrobracon* (Whiting, 1943*a*) and *Apis* (Rothenbuhler, 1957), yield only wild-type 'diploid' males.

Direct cytological evidence on the somatic tissues of *Habrobracon* seems to be lacking. Risler's (1954) study of numerous larval tissues of *Apis*, based partly on chromosome counts and partly on indirect criteria (nuclear and spindle sizes), shows that many male tissues become diploid or polyploid in the later larval stages. But many of these also become polyploid in females, and some tissues, including nerve cells, myoblasts, the antennal imaginal discs and parts of the gut, are still haploid in mature male larvae. O'Brien's (1956) study of *Steatococcus* shows even more strikingly that the situation varies according to the tissue studied. The hypodermis is uniformly haploid in males, diploid in females, both by chromosome counts and DNA content. The fat bodies are mainly haploid in males, but the DNA content is equal to that of female nuclei, presumably as a result of polyteny. In the wax glands the modal DNA contents of male and female nuclei are as 4:2 (!); in the malphigian tubules as 8:32. In *Pteronidea* all the embryonic, larval, and adult tissues examined by Sanderson (1932) were found to be haploid in males, diploid in females, or else polyploid to twice the degree in females as in males. That the ratio between the chromosome number or DNA content of male and female varies according to the species, stage and tissue studied, and is often highly variable even within a single tissue, suggests that somatic polyploidy and polyteny is related to the state of physiological function of the cell, and to cell growth without cell division, not to dosage compensation.

The very widespread and variable occurrence of somatic polyploidy and polyteny in insects and vertebrates generally suggests that most gene functions are relatively little affected by an equal change in dosage of *all* genes simultaneously. So does the fact that balanced genetic polyploids (e.g. triploid *Drosophila* females) are often, apart from cell size, a remarkably close approach to the normal diploid phenotype, whereas aneuploids are usually grossly abnormal or inviable. For characters whose expression depends on the concentration of an intracellular substance (pigmentation being the most obvious example) there is the additional consideration that twice the amount in a cell twice as large will give the same concentration. One may therefore expect that for many, perhaps most, mutant genes, n doses in an n-ploid will give the same effect for different values of n. This is in fact the case for *shaved* and for alleles of *white* in diploid and triploid *Drosophila* females (Schultz, 1935). There are, of course, exceptions; three doses of *cubitus interruptus* in triploid *Drosophila* females give a *less* extreme effect than two doses in diploids (*Stern*, 1943) and, as has been seen, *fused* in *Habrobracon* deviates in the opposite direction.

There clearly is a problem of compensation or adaptation in 'haploid–diploid' species, analogous to the dosage compensation problem for sex-linked genes in other species. The switch between haploidy and diploidy will not automatically give optimally adapted phenotypes; this will evolve only as a result of selection. This will be true in all species in which the males occurring in natural populations are predominantly 'haploid', irrespective of whether 'diploid' males are fully viable, as in *Mormoniella* (Whiting, 1960); of reduced viability, as in *Habrobracon*, or inviable, as in *Apis* (Mackenson, 1955; Rothenbuhler, 1957). The occurrence of such selection (though not perhaps the means by which it becomes effective) will also be independent of the method of sex determination, whether this depends on heterozygosity versus homozygosity for a series of multiple sex 'alleles' (*Habrobracon*; *Apis*) or on some other mechanism (*Mormoniella*). The most obvious 'automatic' (but not unmodifiable) result of haploidy is a reduction of cell size. The cells of the wings and eyes of 'haploid' *Habrobracon* males are almost as large as those of females; those of 'diploid' males are much larger (Speicher, 1935; Grosch, 1945; see also Rothenbuhler, 1957, for 'diploid' male tissue in *Apis*). Whiting (1945) plausibly argues that this is the result of selection for a physiologically optimal cell size in both sexes, this having led to the accumulation of recessive genes for increased cell size in the sex-determining segment. The recessive nature of these genes may of course be subject to modification by other genes which need not themselves lie in the sex-determining segment. Thus (as in other organisms: see the preceding section) the whole genome, not merely the sex-determining segment, will be exposed to bring about such compensatory effects.

Many 'haploid–diploid' species are characterized by very marked sexual dimorphism and, since the 'automatic' results of haploidy will often be neutral, selection will have acted at least as often to *increase* the phenotypic differences between the sexes as to decrease or eliminate selectively disadvantageous 'automatic' consequencies of haploidy. For this reason it may be particularly difficult to infer the existence and nature of such selection from the expression of mutant

genes in 'diploid' males and 'triploid' females; in any case, this has so far received hardly any attention. Somatic diploidy or polyploidy in the male *may* be one of the means by which such selection has been effective, but at present there is little to suggest that it has played a major role. Indeed, to the extent that somatic diploidy does occur, it may be, as Whiting (1945) suggests (see also Suomalainen, 1950), an evolutionary remnant from a time when 'haploid–diploids' originated from forms with tychoparthenogenesis.

5. SUMMARY

1. Inactivation of one X chromosome in somatic cells of female mammals is a form of dosage compensation of sex-linked genes, but the mechanism is entirely different from that operating in *Drosophila*. The latter is designated as dosage compensation *sensu strictu*.

2. There is no dosage compensation of *barred*, *sex-linked dilution* or *slow-feathering* in domestic fowls, of *almond* or *faded* in pigeons, or of *cinnamon* in canaries. Among Lepidoptera the same is true of *sex-linked melanism* in *Lymantria monacha* and of a locus controlling haemolymph colour in *Choritoneura* spp. There is no positive evidence that dosage compensation occurs outside *Drosophila* and mammals.

3. Sex-chromatin in female birds (heterogametic) has been reported by several authors; the genetical evidence is against the possibility that this represents (as in mammals) an inactivated X chromosome. Sex-chromatin in the heterogametic sex also occurs in some (not all) Lepidoptera and Heteroptera; in Heteroptera it usually represents a heteropyknotic Y chromosome.

4. Some complications in Muller's theory of dosage compensation *sensu strictu* are discussed. Not all 'compensatory modifiers' are necessarily sex-linked.

5. The problem of dosage compensation in species with impaternate males is discussed; *fused* in *Habrobracon* is not compensated.

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