Genetica 45: 349–354 *May 12, 1975*

NOTES ON CHROMOSOMES OF TEN SPECIES OF THE GENUS CHRYSOLINA MOTS. (COLEOPTERA: *CHRYSOMELIDAE*)

E. Petitpierre
Departamento de Genética, Universidad de Barcelona, Barcelona, Spain

Received December 4, 1973/Accepted February 1, 1974

Six *Chrysolina* species from Catalonia and the Canary Islands (Spain), viz. *americana, gemina, femoralis, cerealis, menthastri* and *polita*, have similar diploid karyotypes of 24 (sub)metacentric chromosomes, and show Xy sex-determining system. *C. banksi* and *obsoleta* have 2n (♂) = 23; their karyotype is presumably derived from that of the former group by loss of the y chromosomes. In *C. haemoptera* and *C. carnifex* 40 elements appear in the diploid set. It seems that 2n = 24 is the most frequent number in the *Chrysolina*. Higher chromosome numbers have possibly originated through centric fissions, as the acrocentric shape of *C. carnifex* chromosomes seems to suggest. The 2n = 23 and 24 species feed on *Labiatae*, while the two higher chromosome number species are associated with plants belonging to other families.

Introduction

A number of papers have been published on the cytology of *Chrysomelidae* beetles, [Robertson (1966) on the genus *Calligrapha*, Smith (1969) and Ennis (1971) on *Diabroticites*, Virkki (1970, 1971, 1972) on the karyotypes and sex-bivalent mechanisms in *Alticinae*, and Petitpierre (1970) on *Timarcha*], while for a review of the bibliography (1951–1974) on 36 Indian species referable to nine chrysomelid subfamilies reference is made to Yadav & Pillai (1974).

The genus *Chrysolina* has so far not been studied cytologically except for *C. exanthematica*, studied by Yosida [in Smith (1953)]. It includes more than two hundred species, most of which have Palaearctic distribution (Bechyne, 1950).

The aims of our paper are to determine the preliminary chromosomal interrelationships which could help to establish some taxonomic and evolutionary implications characteristic for this genus. A more complete analysis will be actually necessary in the future, in order to obtain a clearer evolutionary picture.

Material and methods

Cytological examinations were performed on spermatogonial cells of adult specimens, collected in the field in autumn 1972 and 1973, and in spring 1973. They were dissected five to ten days after capture.

The slides were stained with Gurr's acetic orcein and metaphase plates were photographed and printed at × 2000. At least two specimens were examined in most of the species.

The species studied are listed in Table 1.

TABLE 1
REVIEW OF THE MAIN CYTOTAXONOMIC DATA OF THE *Chrysolina* SPECIES STUDIED

Species	Source	2n(♂)	Karyotype
americana L.	Garraf (Barcelona)	24	$11^{II} + Xy_p$
gemina Brull.	Tenerife (Canary Isl.)	24	$11^{II} + Xy_p$
femoralis Ol.	La Garriga (Barcelona)	24	$11^{II} + Xy_p$
femoralis Ol.	Collada de Tosses (Pyrenees Gerona)	24	$11^{II} + Xy_p$
femoralis Ol.	Vall. de Llauset (Pyrenees Huesca)	24	$11^{II} + Xy_p$
cerealis L.	Llac Llebreta (Pyrenees Lérida)	24	$11^{II} + Xy_p$
menthastri Suffr.	La Garriga (Barcelona)	24	$11^{II} + Xy_p$
polita L.	La Garriga (Barcelona)	24	$11^{II} + Xy_p$
banksi F.	El Prat (Barcelona)	23	$11^{II} + XO$
obsoleta Brull.	Tenerife (Canary Isl.)	23	$11^{II} + XO$
haemoptera L.	Vall. de Llauset (Pyrenees Huesca)	40	$19^{II} + Xy$
carnifex F.	La Garriga (Barcelona)	40	$19^{II} + Xy_p$
carnifex F.	Aliò (Tarragona)	40	$19^{II} + Xy_p$

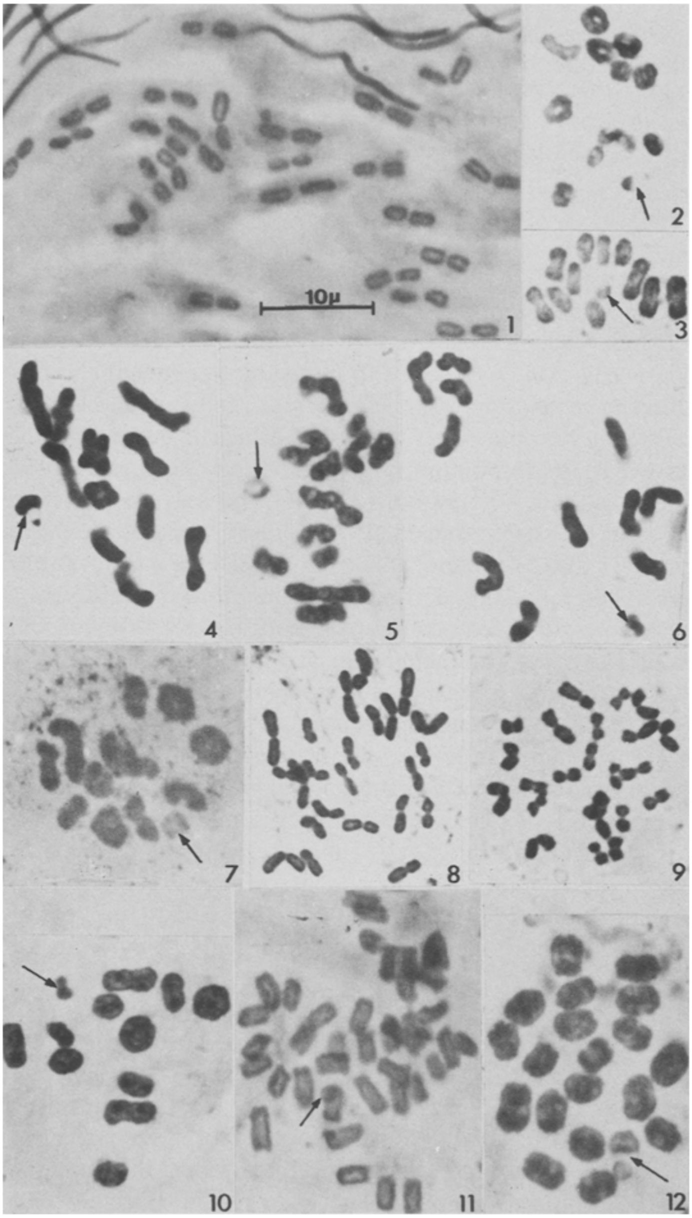
Observations

Chrysolina americana, *gemina*, *femoralis*, *cerealis*, *menthastri* and *polita*, have diploid karyotypes of 24 chromosomes. They consist of large or medium size sub(metacentric) elements which cannot be easily distinguished (Fig. 1). At metaphase I they have eleven rod/round-shaped autosomal bivalents and the typical parachute-like system of Coleoptera sex-chromosomes, Xy_p (Figs. 2–7).

In the spermatogonial metaphases of *C. banksi* and *C. obsoleta* there are 23 (sub)metacentrics of large or medium size (Figs. 8–9). The sex-determining system is of the XO type, as evidenced by the unpaired X-chromosome (Fig. 10).

The only studied specimen of *C. haemoptera* had twenty bivalents at primary metaphase.

C. carnifex has a complement of 40 acrocentrics of medium size, save for a small *y*-chromosome (Fig. 11).



Figs. 1–12. Chromosomes of *Chrysolina* species (Figs. 8–9, ×1500; others ×2000): (1) Spermatogonial metaphase of *C. femoralis*; (2–6) Meiotic metaphases I of *C. americana* (2), *C. gemina* (3), *C. femoralis* (4), *C. cerealis* (5), and *C. menthastri* (6) showing the Xy_p sex-determining system (aceto-orcein squash); (7) Meiotic metaphase I of *C. polita* showing the Xy_p sex-determining system; (8–9) Spermatogonial metaphases of *C. banksi* (8) and *C. obsoleta* (9); (10) Meiotic metaphase I of *C. banksi* with the unpaired X-chromosome; (11) Spermatogonial metaphase of *C. carnifex* showing the y-chromosome indicated by an arrow; (12) Meiotic metaphase I of *C. carnifex* showing the Xy_p sex-determining system (aceto-orcein squash).

At metaphase I there are 20 bivalents. They are uniform in size and larger than in any other representative of the genus. A parachute-like system, Xy_p, can be clearly distinguished at this stage (Fig. 12).

Discussion

The genus *Chrysolina* seems to show a large degree of variation in chromosome numbers which range from 2n = 23 to 2n = 40. Considering our present knowledge, 2n = 24 seems to be the most frequent chromosome number in the genus, since it appears in six of ten analysed species. Other *Chrysomelinae* like the genus *Timarcha*, which is more primitive judged by its external morphology and male genitalia (CHEN, 1934; JOLIVET, 1948; IABLOKOFF–KHNZORIAN, 1966), possess as its more frequent number 2n = 20 (PETITPIERRE, 1970). This formula besides being the most common in the Coleoptera, is also considered the most primitive (SMITH, 1950). Therefore, the chromosomal findings on *Chrysolina* corroborate the more advanced evolutionary position of this genus with regard to that of *Timarcha*, also included in this subfamily.

The six 24-chromosome species of *Chrysolina* are classified into six structurally well defined subgenera (BECHYNE, 1950). It is interesting, therefore, that their cytological features are essentially similar, and could only be distinguished by a careful karyometric analysis, which might make it possible to detect small chromosomal rearrangements. The great cytogenetic uniformity of *C. americana*, *gemina*, *femoralis*, *cerealis*, *menthastri* and *polita*, could only be interpreted as good evidence for their close evolutionary origin, and could also explain possible hybridizations among some of these species, as suggested by JOLIVET (1949).

In the spermatogonial sets of *C. banksi* and *C. obsoleta* there are 23 chromosomes which are morphologically similar to those of the above described species. These two species seem to have evolved by the loss of the *y*-chromosome, changing the sex-mechanism from the primitive Xy_p to the XO system. This change could be understood if the *y*-chromosome consisted of heterochromatic and therefore dispensable material, as has been found in other Coleoptera (DUTRILLAUX, 1970). In addition, the XO sex-system is not rare in *Chrysomelidae* (SMITH, 1950, 1953, 1960 and 1969; ROBERTSON, 1966). The two species are structurally closely allied hence they are classified into the same sub-genus (*Chrysolina* s. str. Mots.).

The last two species studied, viz. *C. haemoptera* and *C. carnifex*, both have 2n = 40. The derivative nature of these karyotypes is remarkably supported in *C. carnifex* by the acrocentric shape of all chromosomes, which may suggest their origin through centric fission. The parachute-like system, Xy_p, is also kept in *C. carnifex* and probably in *C. haemoptera*, but we have no accurate data on this latter species.

The genus *Chrysolina* is divided into thirty-nine subgenera (BECHYNE, 1950). Our ten cytologically analysed species are referable to nine of these though this classification is not reflected by the cytological data.

It is interesting, however, that the host-plant preferences fit well with the cytology since all 23 or 24-chromosome species feed on plants of the family *Labiatae*, while the higher chromosome number species (*C. haemoptera* and *C. carnifex*), live on *Plantago* (*Plantaginaceae*) and *Artemisia* (*Compositae*) respectively. Consequently, the *Chrysolina* adaptation to host-plants seems to have been accompanied by great chromosomal rearrangements, through increases in chromosome number. These chromosomal increases have also been observed in other *Chrysomelinae* like the genus *Timarcha*, although they are not correlated with new plant adaptations (PETITPIERRE, 1969, 1970). On the other hand, small chromosomal changes like the *y*-chromosome loss, or undetected other rearrangements, could accompany the process of speciation in the cytologically similar *Chrysolina*, providing the suitable genetic constitution to occupy new ecological habitats, with sometimes specific plants of *Labiatae*.

I am indebted to Prof. A. PREVOSTI for advice and critically reading the manuscript and to Mr. P. OROMI for providing individuals of the two species from the Canary Islands.

References

BECHYNE, J. (1950). 7e contribution à la connaissance du genre Chrysolina Mots. (Col., Phyt., Chrysomelidae). *Ent. Arb. Mus. Georg Frey, München,* **1:** 47–185.

CHEN, S. H. (1934). Sur la position systématique du genre Timarcha Latr. *Bull. Soc. ent.* Fr. **39:** 35–38.

DUTRILLAUX, B. (1970). Étude cytogénétique de quatre espèces de Prioniens (Col. Cerambycidae). *Annls Soc. ent. Fr.* (N.S.) **6:** 443–450.

ENNIS, T. J. (1971). Meiosis in Diabrotica (Coleoptera: Chrysomelidae): Chiasma frequency and variation. *Can. J. Genet. Cytol.* **14:** 113–128.

IABLOKOFF-KHNZORIAN, S. M. (1966). Considérations sur l'adéage des Chrysomelidae et son importance phylogénique. *Entomologiste* **22:** 115–138.

JOLIVET, P. (1948). Introduction à la biologie des Timarcha (Col. Chrysomelidae). *Miscnea ent.* **45:** 1–32.

JOLIVET, P. (1949). Les hybrides d'espèces chez les Chrysolina Mots. (Col. Chrys.). *Entomologiste* **5:** 77–80.

PETITPIERRE, E. (1969). Contribuciones citogenéticas a la filogenia de las Timarcha (Col. Chrysomelidae). *I Simp. Int. Zoofilogenia, Salamanca,* pp. 395–406.

PETITPIERRE, E. (1970). Cytotaxonomy and evolution of Timarcha Latr. (Coleoptera: Chrysomelidae). *Genet. iber.* **22:** 67–120.

ROBERTSON, J. G. (1966). The chromosomes of bisexual and parthenogenetic species of Calligrapha (Coleoptera: Chrysomelidae) with notes on sex-ratio, abundance and egg number. *Can. J. Genet. Cytol.* **8:** 695–732.

SMITH, S. G. (1950). The cyto-taxonomy of Coleoptera. *Can. Ent.* **82:** 58–68.

SMITH, S. G. (1953). Chromosome numbers of Coleoptera. *Heredity* **7:** 31–48.

SMITH, S. G. (1960). Chromosome numbers of Coleoptera II. *Can. J. Genet. Cytol.* **2:** 66–88.

SMITH, S. G. (1969). The chromosomes of some chrysomelid Coleoptera: Diabroticites. *Chromosomes Today* **3:** 197–207.

VIRKKI, N. (1970). Sex chromosomes and karyotypes of Alticidae (Coleoptera). *Hereditas* **64:** 267–282.

VIRKKI, N. (1971). Formation and maintenance of the distance sex bivalent in Oedionychina (Coleoptera: Alticidae). *Hereditas* **68:** 305–312.

VIRKKI, N. (1972). Contraction stage and formation of the distance sex bivalent in Oedionychina (Coleoptera: Alticidae). *Hereditas* **71:** 259–288.

YADAV, J. S. & R. K. PILLAI (1974). Chromosome numbers of Indian Coleoptera. *Genetica* (in press).