

Fig. 3. Karyotype of Carollia perspicillata.

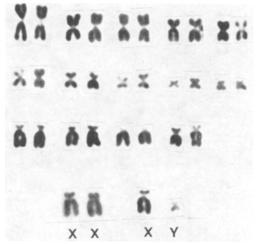


Fig. 4. Karyotype of Sturnira lilium.

The results of our investigations do not show any significant difference with the karyotypes described for the same species from various localities of Mexico, as reported by Baker¹ and by Hsu et al.², separated by more than 3000 km. The same is true for A. lituratus as compared with the information afforded by Beçak et al.6 from specimens of São Paulo, Brazil.

The double Y sexual system in males, found in very few mammalian species, would seem to be a peculiar characteristic of some species of leaf-nosed bats, and it is found in at least 2 different subfamilies (Phyllostominae and Carolliinae). The origin of these allosomes is apparently different, since their shapes are markedly different in the 2 subfamilies. At any rate, phylogeny of these allosomes is still doubtful and new information will be needed to shed light on this problem and to determine its true extension and significance within the group.

Resumen. Se estudiaron los cromosomas de 7 especies de murciélagos filostómidos de Venezuela. Se compararon con los estudios realizados en México, no encontrándose diferencias significativas y confirmando el sistema XY_1Y_2 en algunas especies. En $Ph.\ hastatus$ se observó una diferencia en un par autosómico; un caso de variación intragenérica.

P. Kiblisky⁷

Instituto de Zoologia Tropical, Facultad de Ciencias, Universidad Central de Venezuela Caracas (Venezuela), 6 May 1969.

- 6 M. L. BEÇAK, R. F. BATISTIC, L. D. VIZOTTO and W. BEKAC, Experientia 25, 81 (1969).
- I thank O. A. Reig for constructive criticism and encouragement, J. R. Tamsitt, O. Linares, P. Soriano and M. Tuttle who collected the material discussed in this paper, E. Garcia for correcting the manuscript. J. R. Tamsitt and O. Linares are responsible for zoological identifications.

Nucleolonema in Lemon Fruit Nucleoli (Citrus limon L.)

The heterogeneous nature of the nucleolus has been demonstrated in plant and animal organisms. Opinions differ, however, concerning the structural components of the nucleolus, especially with respect to the nucleolonema-nucleoloplasm concept of Estable and Sotelo¹⁻⁵. Optical confirmation of the presence of the nucleolonemata in nucleoli of various plant species has been obtained recently ^{6,7}. This communication presents evidence of the presence of a nucleolonema in interphase nuclei of lemon fruit tissue, a structure which has previously been referred to as 'strand-like' and 'linear' components ⁸⁻¹¹.

Materials and methods. Stalks from juice vesicles of mature lemon fruits (Citrus limon L.) were inoculated aseptically onto mineral-sucrose solution (pH 5.0–6.0) in 'Pyrex' Petri dishes lined with Whatman No. 40 or 42 'Ashless' filter paper or Whatman GF/A glass filter paper and kept in the dark at 25 °C1². 1–3-day-old explants were fixed in Randolph's CRAF solution ¹³ and unstained paraffin sections (12 μ thick) and unstained squash pre-

parations made as described previously ^{10, 11, 14}. Squash preparations dehydrated with isopropanol were cleared with xylene and mounted in 'Sira' mountant instead of 'Euparal'.

- ¹ J. L. Sirlin, Prog. Biophys. biophys. Chem. 12, 25 and 319 (1962).
- ² B. A. Kihlman, Actions of Chemicals on Dividing Cells (Prentice-Hall Inc., New Jersey 1966), p. 38.
- ³ C. H. WADDINGTON, Natn. Cancer Inst. Monograph 23, 563 and 573 (1966).
- ⁴ M. Birnstiel, A. Rev. Plant Physiol. 18, 25 (1967).
- ⁵ C. Estable and R. J. Sotelo, Symposium on Fine Structure of Cells (P. Noordhoff Ltd., Publishers, Groningen 1955), p. 170.
- ⁶ L. F. LA COUR, *Chromosomes Today* (Eds. C. D. DARLINGTON and K. R. LEWIS; Oliver and Boyd; Edinburgh and London 1966), vol. I, p. 150.
- ⁷ A. LORD and J. G. LAFONTAINE, J. Cell Biol. 40, 633 (1969).

Results and discussion. Nucleoli of interphase nuclei of the paraffin-sectioned and squashed explants frequently contained material that was very refractile under phase contrast microscopy. In the sectioned tissue this refractile nucleolar material appeared at times as filamentous structures and at times as 'granules' depending upon its spatial orientation within the nucleoli. Squash preparations, however, consistently revealed the presence of a definitive filamentous nucleolar component (Figures 1, 2, 3a, 3b) which oftentimes looked like the nucleolonemata described by LA COUR⁶ (compare Figure 1 with Figure d of Plate II of reference number ⁶). Compression of the refractile nucleolar material into few optical planes

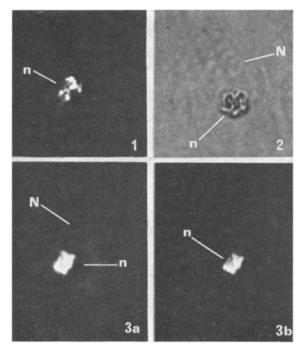


Fig. 1. Unstained squash preparation showing the prominent and highly refractile nucleolonema in the nucleolus. (See Figure d, Plate II of reference number ⁶.) Phase contrast. × 1350.

Fig. 2. Unstained squash preparation which clearly shows the filamentous nature of the nucleolonema. Bright field microscopy using a Balzers green interference filter (547 nm). \times 1350.

Figs. 3a, 3b. Unstained squash preparation showing the highly refractile nucleolonema in the shape of a rectangle. Note that the fillamentous nature of the nucleolonema in 3a is obscure and the nucleus and nucleolus are visible whereas in 3b the fillamentous nature of the nucleolonema is apparent and the nucleus and nucleolus are no longer visible (see footnote ¹² of reference number ¹⁴). Phase contrast. × 1350. N, nucleus; n, nucleolus.

during squashing was most likely a factor responsible for the more definitive optical resolution of its filamentous nature in the squash preparations than in the sectioned material. Physical displacement of this nucleolar component in the squashed tissue also served to substantiate its filamentous nature since its linear structure was evident regardless of the varied shapes that it assumed during squashing.

Fabbri 15 has suggested that nucleoli of plant cells contain granules which may give the appearance of being filamentous structures because of optical artifacts. ESTABLE and Sotelo⁵ consider nucleolar inclusions such as granules, vacuoles, or networks described by other investigators as being optical misinterpretations of the filamentous nucleolar component they call the nucleolonema. Figures 3a and 3b shown here are of particular interest in that the filamentous nucleolar component has assumed a rectangular appearance following squashing, a shape not likely to be formed from random orientations of granules or vacuoles. The persistent linearity within the refractile nucleolar component irrespective of the overall shape assumed under the physical pressure of squashing likewise would not be expected if this nucleolar component consisted of randomly-situated inclusions such as granules or vacuoles instead of a filamentous structure.

The observations described here do not preclude the existence of various kinds of nucleolar inclusions described by other investigators^{3,4}. They do, however, confirm the presence of a filamentous component, the nucleolonema^{5-7,16}, in nucleoli of interphase nuclei of lemon fruit tissue.

Zusammenfassung. Die Anwesenheit eines Nukleolonemas in den Zwischenphasennukleolen des Zitronenfruchtgewebes wird bestätigt.

H. A. KORDAN

Department of Botany, University of Birmingham, Birmingham 15 (England), 14 July 1969.

- 8 H. A. Kordan and L. Morgenstern, Expl. Cell Res. 28, 133 (1962).
- 9 H. A. KORDAN and L. MORGENSTERN, Expl. Cell Res. 30, 98 (1963).
- ¹⁰ H. A. Kordan and R. D. Preston, Nature 216, 1105 (1967).
- ¹¹ H. A. Kordan, Experientia 25, 743 (1969).
- 12 H. A. KORDAN, Phyton 26, 31 (1969).
- ¹⁸ D. A. Johansen, *Plant Microtechnique* (McGraw-Hill Book Co. Inc., New York and London 1940), p. 45.
- ¹⁴ H. A. Kordan, Experientia 25, 517 (1969).
- ¹⁵ F. Fabbri, Caryologia 16, 715 (1963).
- 16 C. Estable, Natn. Cancer Inst. Monograph 23, 91 (1966).

In vitro Induction of Flowering in Cucumis sativus L.

Sex expression in cucumber has been studied by using either whole plants in vivo^{1,2} or isolated floral buds in vitro³. Recently, we have found that isolated shoot apices of this plant can be grown into plants which produce flowers in vitro. This technique should prove very useful in studying the effects of growth regulating chemicals and environmental factors on sex expression of cucumber.

Seeds of *Cucumis sativus* L. var. 'Long Green Improved', a monoecious variety, were obtained from a commercial seed supply house. They were sterilized in a

¹ C. E. Petersen and F. J. Kribben, Science 131, 1673 (1960).

² D. Atsmon, A. Lang and E. N. Light, Plant Physiol. 43, 806 (1968).

³ E. Galun, Y. Jung and A. Lang, Dev. Biol. 6, 370 (1963).