Einfluß von Ionenstärke und pH auf die differentielle Dekondensation der Nukleoproteide isolierter Speicheldrüsen-Zellkerne und -Chromosomen von Chironomus thummi

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Einfluß von Ionenstärke und pH auf die differentielle Dekondensation der Nukleoproteide isolierter Speicheldrüsen-Zellkerne und -Chromosomen von Chironomus thummi

MICHEL ROBERT
Zoologisches Institut der Eidgenössischen Technischen Hochschule, Zürich, und Institut für Genetik der Universität des Saarlandes, Saarbrücken

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Effect of Ionic Strength and pH on the Differential Decondensation of Nucleoproteins in Isolated Salivary Gland Nuclei and Chromosomes of Chironomus thummi

Abstract. A new method of isolating nuclei and chromosomes of salivary gland cells is described. — The influence of ionic strength and pH of the medium on the state of decondensation of chromosomal bands is studied. In the isolation medium (a modified Ringer solution), all the bands are in a condensed state; as the ionic strength is increased the bands decondense. This reaction of the bands to increasing ionic strength is dependent on the pH which determines: 1) the range of ionic strengths which causes decondensation of the bands; i.e., the lower the pH, the higher the ionic strength is required for decondensation (at pH 7.3, 150–350 mM NaCl, at pH 4.3, 500–800 mM NaCl), and 2) the extent of structural changes caused by increasing ionic strength; that is, at neutral pH the bands become diffuse ("fading") and at moderate acidic pH (optimum 4.3) the bands unravel to yield puff-like structures ("swelling"). — All ion species tested induce decondensation of bands, but each one is effective differently; specifically, Mg²⁺ is more effective than Na⁺ and K⁺, and ClO₄⁻ is more effective than Cl⁻. — "Swelling" as induced at pH 4.3 by high ionic strength cannot be reversed by a mere lowering of ionic strength (to 150 mM NaCl) and a subsequent raise of pH (to 7.5); it can be reversed only by an addition of histones. The various histone fractions act differently on the recondensation process. — "Swelling" is correlated with an increase in template activity as evidenced by an increased incorporation of ³H-UTP, measured in the presence of ATP, CTP, GTP and exogeneous RNA polymerase. — The individual bands differ in their sensitivity to an increasing ionic strength. This differential sensitivity expresses itself only if one of the following conditions is met: 1) a moderately acidic pH (optimum 4.3) or 2) the presence of divalent cations at neutral pH. — In a few bands the sensitivity to an increasing ionic strength is dependent on the ionic species (Na⁺, K⁺, Mg²⁺ and Ca²⁺). — It is attempted to explain the above reactions on the basis of the physico-chemical properties of chromosomes (Zusammenfassung see p. 30).

A. Einleitung

In Interphase-Zellkernen höherer Organismen kann das Chromatin kondensiert und inaktiv oder dekondensiert und aktiv vorliegen. Beide