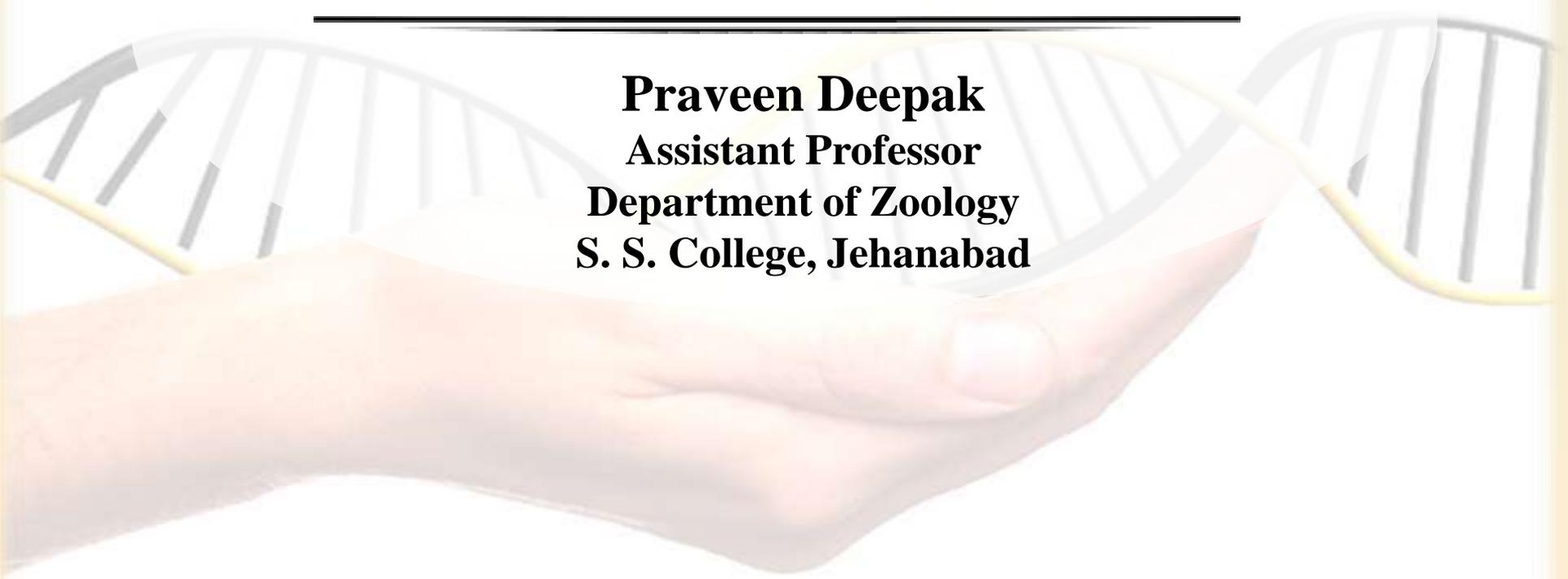


Structural organization and function of polytene chromosome

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Introduction

- ❑ Polytene chromosomes are giant interphase chromosomes with several DNA strands placed side by side.
- ❑ They are first observed by E. G. Balbiani in 1881 in squash of salivary gland cells of Chironomous larvae, hence also known as salivary gland chromosome or giant chromosome.
- ❑ They are strongly amplified form of interphase chromosomes that are commonly seen in salivary glands of dipteran species, e.g. *Chironomus* larva, *Drosophila melanogaster* larvae, *Sciara*, *Rhyncosciara*, etc.
- ❑ They are also found in various other tissue in the larvae of dipteran, such as midgut, rectal and Malpighian tubules as well as in several species of protozoans and plants.
- ❑ They are many times larger than the normal chromosomes reaching a length of $200\mu\text{m}$ and are visible even under a compound microscope.
- ❑ The enormous size is due to the duplication of chromonema which do not separate.
- ❑ Non-separation of chromonema results in presence of many strands and therefore, it is known as polytene chromosome.
- ❑ Sutton worked on polytene chromosomes of *Anopheles* mosquitoes.
- ❑ Riandian worked on polytene chromosomes of Tse-tse fly.

Flies with polytene chromosome



Adult Lake fly (*Chironomus sp.*)



Adult Fruit fly (*Drosophila melanogaster*)



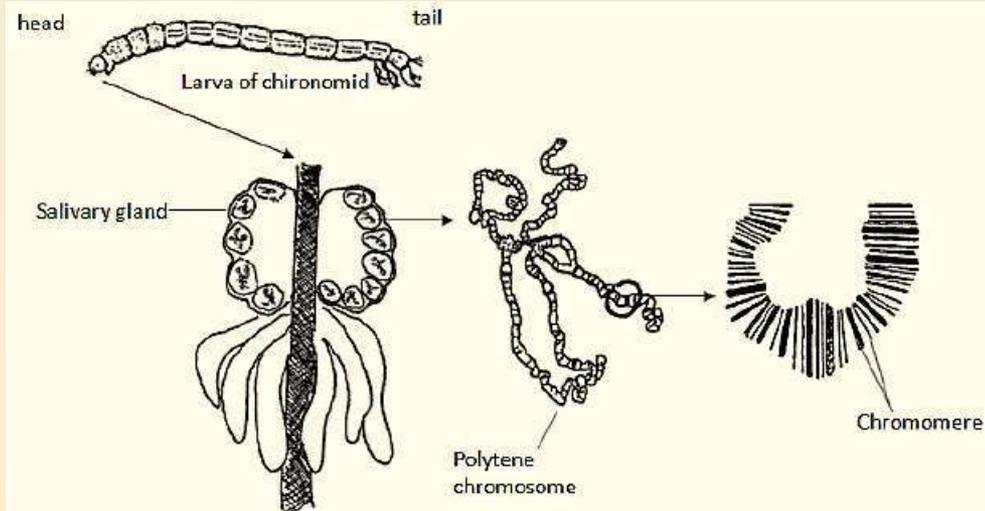
Chironomus larvae



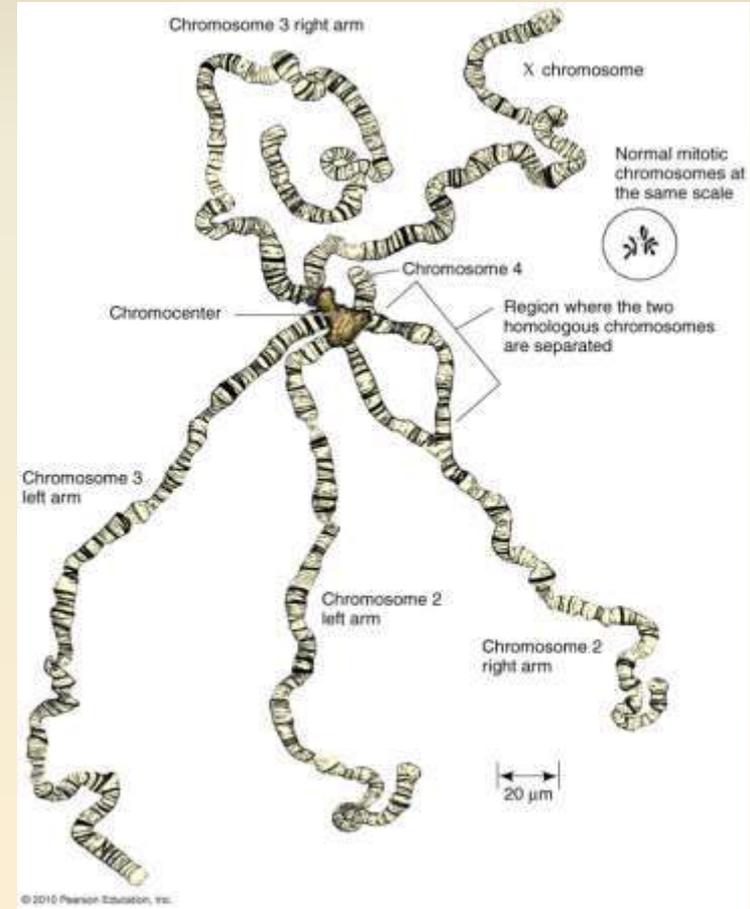
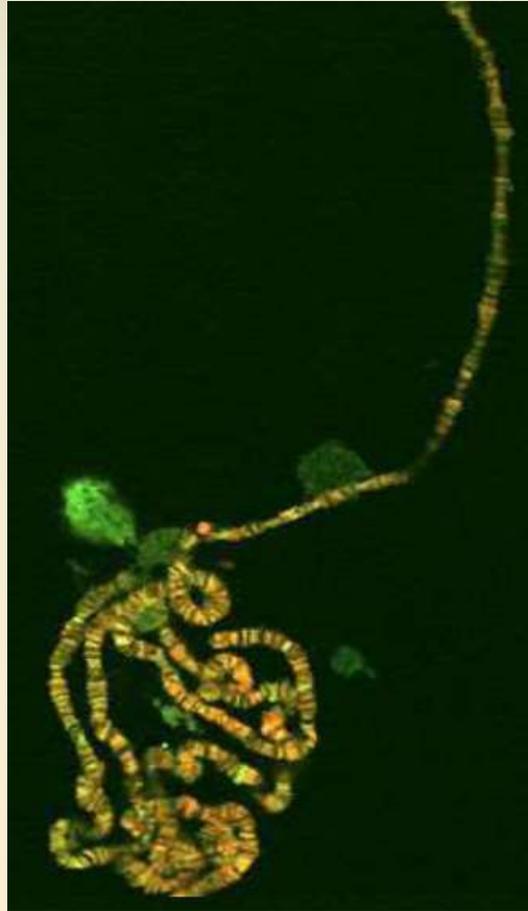
Drosophila melanogaster larva

Morphology of polytene chromosome

- ❑ It is seen in the nucleus during interphase.
- ❑ It is a giant chromosome with 5 long and 2 short arm radiating from a central point called chromocentre. It is 2000 μm in length (7.5 μm in somatic cells).
- ❑ Chromocentre is formed by the fusion of centromeres of all the chromosome ($2n = 8$ in *Chironomus* & $2n = 4$ in *Drosophila*) found in the cell.
- ❑ Out of 6 arms, the short arm represents the fused IV chromosome.
- ❑ The II chromosome is the longest and is metacentric, with seven weak spots.
- ❑ The longest represent the fused sex chromosome.
- ❑ It shows linear series of alternating bands and interbands which are species specific.



Morphology of polytene chromosome



Morphology of polytene chromosome

- ❑ It has 6 radiating arms from chromocentre (5 long and 1 short arm) as described below:
 - I. X-chromosome (longest arm) (Y chromosome is fused with chromocentre in male and usually not seen as separate strand)
 - II. 2R (II-chromosome Right arm)
 - III. 2L (II-chromosome Left arm)
 - IV. 3R (III-chromosome Right arm)
 - V. 3L (III-chromosome Left arm)
 - VI. IV-chromosome (Shortest arm)
- ❑ Chromatin of polytene chromosomes are stained in banded pattern. The chromatin in darkly stained band is more condensed than lightly stained interbands.
- ❑ The dark bands represents heterochromatin region which have no gene or permanently repressed genes, while lightly stained regions contain genes (euchromatin)
- ❑ About 80% of the DNA is located in bands & about 15% in interbands, that means only 15% of DNA is genetically active.
- ❑ It was Painter (1933) & Bridges (1936), who showed that in *Drosophila* the bands are associated with genes.
- ❑ In *Drosophila*, 5000 bands have been found in the 4 chromosomes of salivary gland cells. It was established by Bridges in ~1936 by constructing salivary gland chromosome map of *D. melanogaster*.
- ❑ Dark bands have a high DNA content & absorb U.V light.

Why it is so giant?

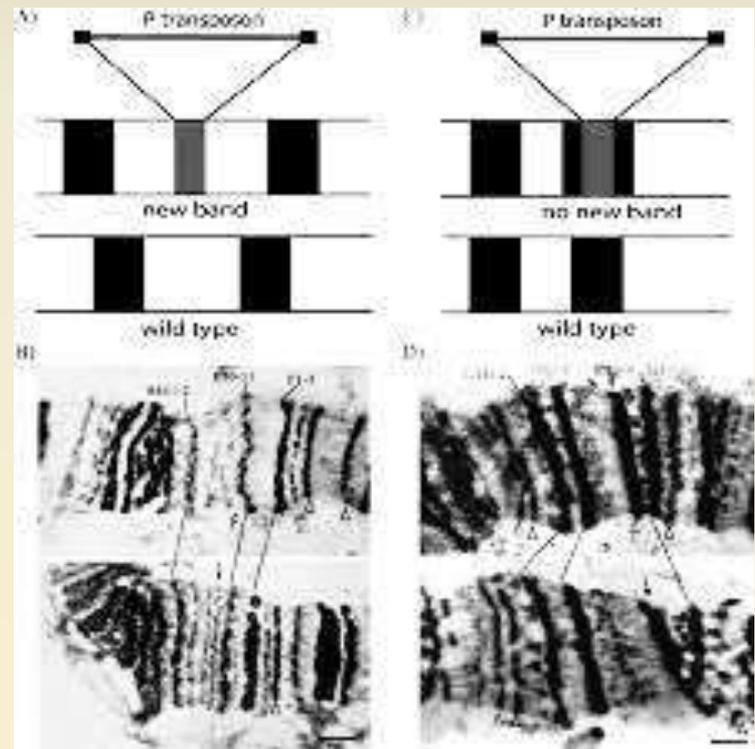
- ❑ It is 1000 times larger than other somatic chromosomes and as long as 0.5mm, and up to 20mm in diameter looking giant.
- ❑ The larger size of the chromosomes is due to the presence of many longitudinal strands called chromonemata.
- ❑ Hence they are also called Polytene chromosomes (many threaded).
- ❑ The many strands of the giant chromosomes are due to repeated division of the chromosome without the cytoplasmic division/mitosis. This type division is called endoreduplication or endoreplication.
- ❑ *Drosophila* chromosomes has been replicated through 10 cycles without separation of the daughter chromosomes, so that $1024 (= 2^{10})$ identical strands of chromatin are lined up side by side.
- ❑ Polytene homologues are held together by somatic pairing.
- ❑ The centromeres of all four chromosomes aggregate to form a chromocentre that is seen amorphous and consists largely of heterochromatin.

Polytenization

- ❑ It is the process of making multiple copies of chromosomes without the segregation and cell division.
- ❑ It occurs when the DNA repeatedly replicates, but the daughter chromosomes do not separate – no karyokinesis.
- ❑ The result is an enlarged chromosome composed of many parallel copies of itself .
- ❑ The amplification of chromosomal DNA greatly increases gene copy number, presumably to supply sufficient mRNA for protein synthesis in the massive salivary gland cells.

Banding pattern of polytene chromosomes

- ❑ The Polytene chromosome contains two types of transverse bands, namely dark bands and inter bands.
- ❑ The dark bands are darkly stained and the inter bands are lightly stained with nuclear stains.
- ❑ The dark bands contain more DNA and less RNA.
- ❑ The inter bands contain more RNA and less DNA.
- ❑ Since the bands can be recognized by their different thicknesses and spacings, each one has been given a number to generate a polytene chromosome "map."
- ❑ The bands in *Drosophila polytene chromosomes* represent 50,000–100,000 base pairs.
- ❑ There are approximately 5000 bands in the total *Drosophila* genome.



Functional stages in polytene chromosome

- ❑ Polytene chromosomes form structures that correlate with the functional state.
- ❑ During the larval development of drosophila, a series of expansions (puffs) appear in temporal stages in the polytene chromosomes.
- ❑ Chromosome puffs are decondensed, expanded segments that represent active chromosomal regions, i.e., regions that are being transcribed.
- ❑ The location and duration of the puffs reflect different stages of larval development.
- ❑ The incorporation of radioactively labeled RNA has been used to demonstrate that RNA synthesis, a sign of gene activity (transcription), occurs in these regions

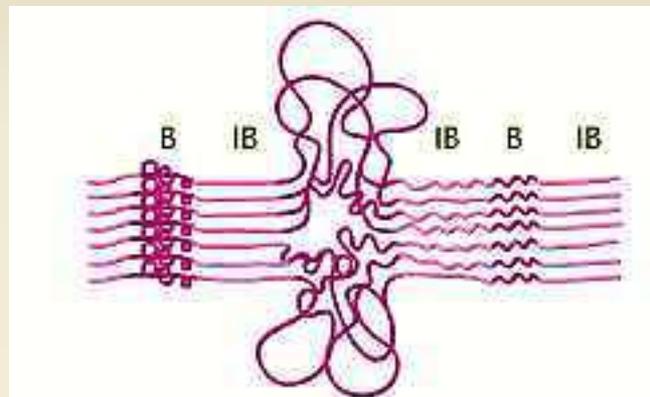
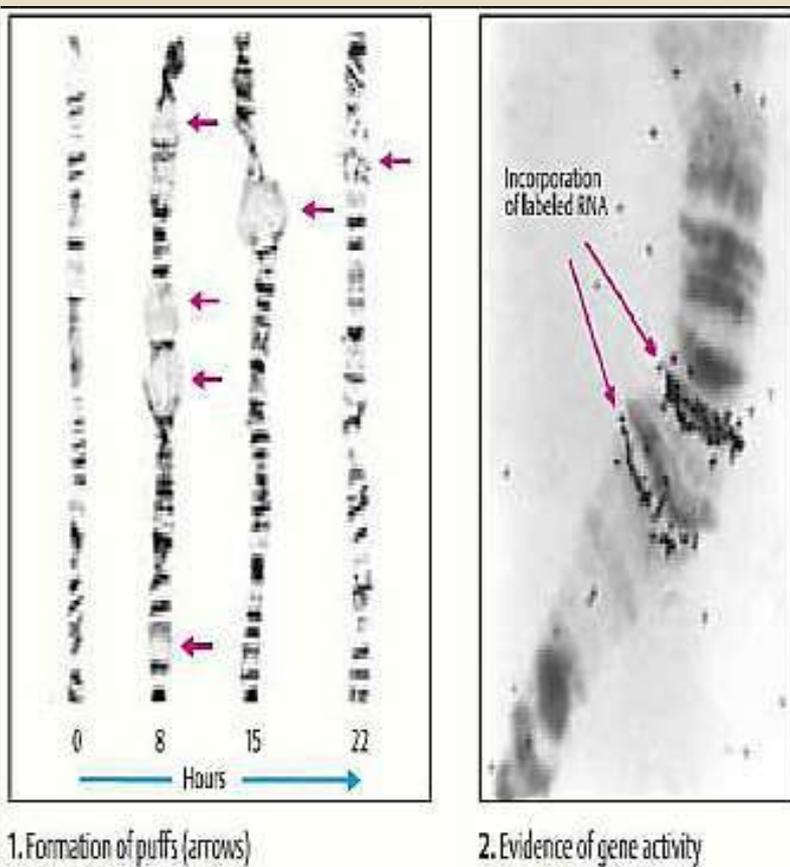
Chromosomal puffs or Balbiani Rings

- ❑ The bands of Polytene chromosomes become enlarged at certain times to form swellings called puffs or Balbiani rings. The formation of puffs is called puffing.
- ❑ In the regions of puffs, the chromonemata uncoil and open out from many loops. Thus puffing is caused by the uncoiling of individual chromomeres in a band.
- ❑ The puffs indicate the site of active genes when mRNA synthesis takes place.



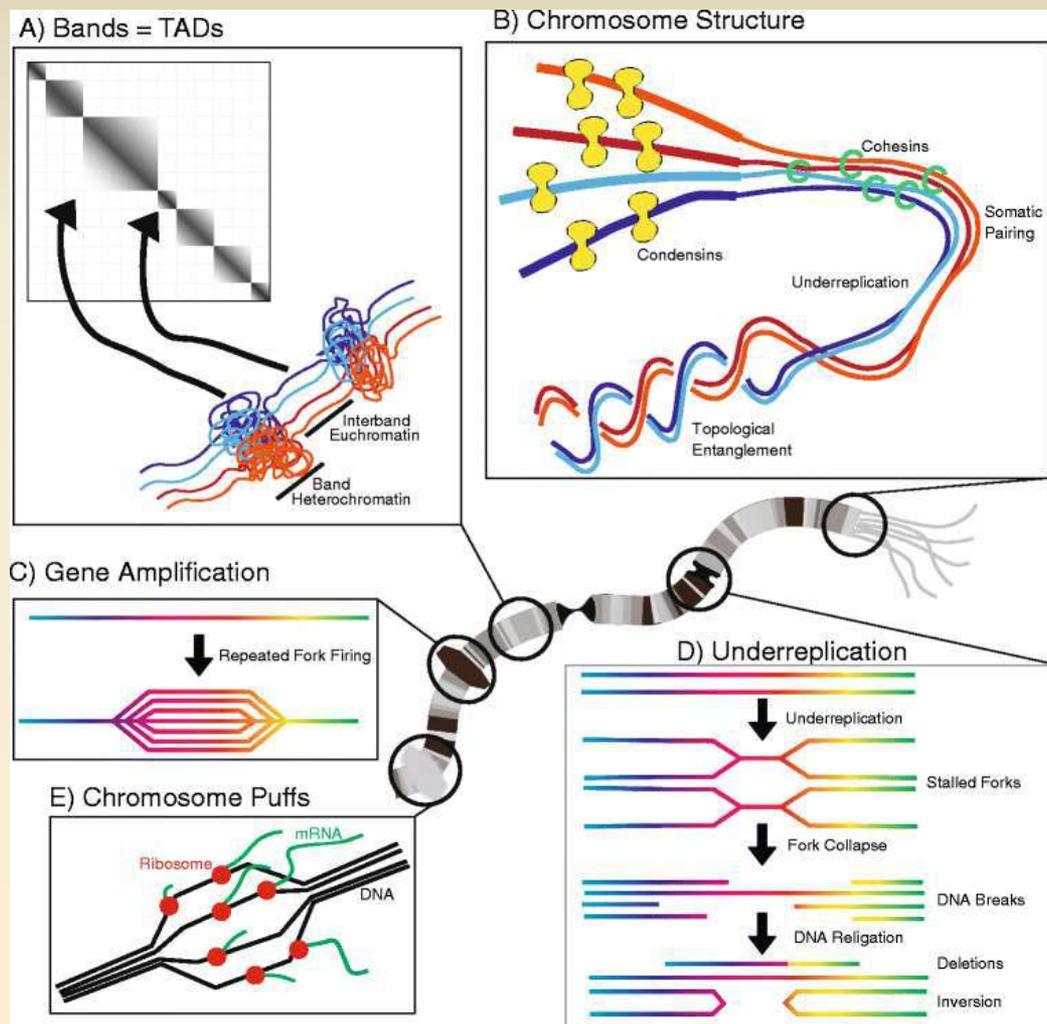
- ❑ Several factors hold sister chromatids together, including topological entanglement caused by DNA coiling, underreplication in some species and cell types, somatic pairing in Diptera and possibly in some ciliates, and cohesin complexes.
- ❑ In order to separate polytene chromosomes into individual strands, condensins are required.

Chromosomal puffs or Balbiani Rings



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Chromosomal puffs or Balbiani Rings



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Function of polytene chromosomes

- ❑ Increasing the volume of the cells' nuclei and causing cell expansion.
- ❑ Metabolic advantage as multiple copies of genes permits a high level of gene expression.
- ❑ In *Drosophila melanogaster*, the chromosomes undergo many rounds of endoreduplication to produce large amounts of glue before pupation.
- ❑ There is tandem duplication of various polytene bands located near the centromere of the X chromosome which results in the Bar phenotype of kidney-shaped eyes.
- ❑ Because polytene chromosomes are interphase chromosomes, and are therefore transcribed. Thus it provides opportunity to study transcription by direct observation, and transcriptional responses to specific stimuli can be observed.

Further reading

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