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A Drosophila melanogaster H3.3 cDNA encodes a histone variant identical with the vertebrate H3.3

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Brief Notes

A *Drosophila melanogaster H3.3* cDNA encodes a histone variant identical with the vertebrate H3.3*

(Conserved proteins; amino acid sequence homologies; chromatin; bacteriophage λ library; invertebrate)

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SUMMARY

A cDNA encoding an H3.3 histone variant in *Drosophila melanogaster* predicts a protein with an amino acid (aa) sequence identical with that in vertebrates. The *D. melanogaster H3.3* nucleotide (nt) sequence has diverged significantly from that of both the *H3.3* gene of vertebrates and the *H3.1* gene of *D. melanogaster*, largely through third nt changes in its codons. The perfect H3.3 aa sequence conservation between organisms as phylogenetically divergent as vertebrates and flies suggests that the H3.3 histone variant itself is an important structural component of chromatin, apart from the value of its replication-independent expression pattern.

The genes encoding H3 in vertebrates include both those expressed under cell-cycle regulation (i.e., *H3.1* and *H3.2* genes) and those expressed under replication-independent control, notably *H3.3*. The latter genes are characterized as containing introns and producing a polyadenylated message, in contrast to the former, which lack introns and produce a nonpolyadenylated message (for review of data see Wells et al., 1989; Wells and McBride, 1989).

We have recovered a clone encoding histone variant H3.3 from a library of *D. melanogaster* embryo cDNA (Brown and Kafatos, 1988) screened with a 673-bp insert of the chicken *H3.3B* clone (Brush et al., 1985). Both strands of the *D. melanogaster* cDNA clone were fully sequenced. *D. melanogaster* H3.3 shows 96% aa sequence similarity to the presumed replication-dependent H3.1 protein of *D. melanogaster* (five single aa changes). The nt sequence, however, displays only 77% similarity to the *D. melanogaster H3.1* gene from the repetitious cluster at polytene chromosome locus 39D2-E2 (Matsuo and Yamazaki, 1989), containing numerous silent nt substitutions. cDNA and genomic clones encoding the H3.3 replication-independent histone variant have also been isolated from human, chick, rabbit, and mouse (Brush et al., 1985; Wells and Kedes, 1985; Wellman et al., 1987; Wells et al., 1987; Hraba-Renevey and Kress, 1989; Chalmers and Wells, 1990). Although the interspecies nt sequences are only 75% similar, the encoded H3.3 proteins are identical.

The H3.3 proteins from human, chick, rabbit, mouse and *D. melanogaster* all differ from their respective H3.1s in having Ser (rather than Ala^{31}), and Ala-Ala-Ile-Gly (rather than Ser^{87}-Ala-Val-Met^{90}). Each variant contains about 75% similarity to its own S-phase-regulated *H3*. Although each organism exhibits numerous silent nt changes while conserving the H3.3 aa sequence, these silent nt substitutions are different among various organisms. The characteristic high degree of similarity present between the vertebrate 3′-untranslated regions (Wells et al., 1987; Hraba-

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* On request the authors will supply experimental evidence for the conclusions reached in this Brief Note.

Abbreviations: aa, amino acid(s); bp, base pair(s); *D.*, *Drosophila*; H3, histone H3; *H3*, gene (DNA) encoding H3; nt, nucleotide(s).

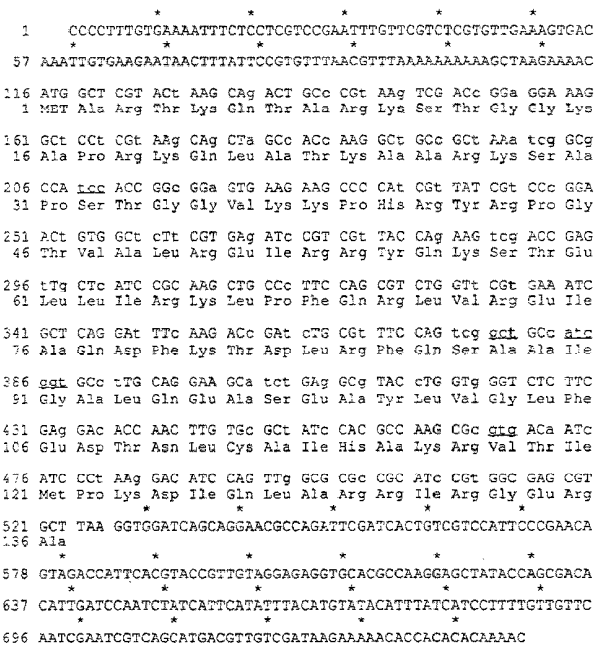

Fig. 1. The nt sequence of a *D. melanogaster* cDNA encoding histone H3.3. Lower-case letters within the coding region indicates a nt substitutions in comparison to the *D. melanogaster H3.1* gene; underlining indicates an aa substitutions. If the cDNA represents a full-length transcript, then the transcription start point is at the 5′ end of the sequence. No TATA box and no polyadenylation signal consensus sequences are present. EMBL accession No. X53822.

Renevey and Kress, 1989; Chambers and Wells, 1990) is not seen with the *D. melanogaster H3.3* cDNA. The function of the H3.3 variants remains unknown; however, the high degree of aa sequence conservation observed argues that the selective pressures on H3.3 reflect the use of the protein itself, and not only a requirement for replication-independent H3 synthesis. Genetic studies in *D. melanogaster* should help elucidate the importance of both the H3.3 histone variant itself and its expression pattern.

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