

# This Week's Citation Classic™

Bonner J, Dahmus M E, Fambrough D, Huang R C, Marushige K & Tuan D Y H.

The biology of isolated chromatin. *Science* 159:47-56, 1968. [Div. Biol., Calif. Inst. Technol., Pasadena, CA; Dept. Biol., Johns Hopkins Univ., Baltimore, MD; Dept. Biochem., Univ. British Columbia, Vancouver, Canada; and Dept. Chem., Harvard Univ., Cambridge, MA]

This paper is a comprehensive review of chromatin biology and chemistry as seen in 1968, or eight years after the beginning of this science. Chromatin consists of DNA complexed with proteins known as histones. It is shown that histones are comprised of five species which are very much the same in all eukaryotic creatures and conserved in amino acid sequence to a remarkable degree, that all five histones are present in all organs of the same creature, and so forth. It is also shown that the complexing of histones to DNA in general makes the DNA a template for transcription of RNA. The paper also concerns the dissociation of chromatin into its constituent proteins and DNA and the reconstitution of the proteins and DNA into chromatin by gradient dialysis. [The *SCI*® indicates that this paper has been cited in over 445 publications since 1968.]

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"I began work on the structure of chromatin in 1960, together with Ru-chih C. Huang.<sup>1,2</sup> In very short order we found that chromatin from eukaryotic organisms is readily prepared, we established criteria of purity, and we discovered RNA polymerase and partially purified it from chromatin of eukaryotic chromosomes of pea plants and rats. At that time, the field of study of isolated chromatin did not exist although some work had been done by Alfred E. Mirsky,<sup>3</sup> but little had been discovered about chromosomal chemistry or structure. This review paper summarizes some of our early findings, namely, that chromatin is DNA complexed with a typically chromosomal class of proteins, the histones; that chromatin serves as a very poor template for DNA synthesis by DNA-dependent RNA polymerase prepared from chromosomes, but that deproteinized DNA serves as an excellent template. Thus, one can imagine that the histones are interfering with the role of DNA as a template for RNA synthesis. We also showed in this paper that genes which are expressed in an organ are expressed in the chromatin of that organ, in the following sense. We prepared

chromatin from pea plant apical buds and from pea plant cotyledons, and gave each RNA polymerase and a cell-free protein synthesizing system prepared from *E. coli* together with all of the essential amino acids, ATP, and the riboside triphosphates. This was then a chromosomally supported messenger RNA generating system, coupled to a protein translation system. We found that in the coupled system, chromatin of pea cotyledons makes messenger RNA that supports the formation of pea seed globulin, while chromatin from pea buds, which do not make pea seed globulin, does not form RNA which supports the synthesis of this protein. A very elegant experiment.

"Although the histones had been known for almost 100 years, their chemistry was quite obscure, and Huang and I decided to "grow" our own histone chemist, in the person of Douglas Fambrough. Fambrough, by early 1968, had made order out of chaos. He had shown that there are five species of histone molecules in both pea chromatin and calf thymus chromatin, and that the histones of the analogous classes from the two kinds of chromatin are very similar to one another. Later in 1968, we found by the sequencing of histone IV, the smallest and most readily purifiable of the histones, that the histones IV of pea and cow are essentially identical, each 102 amino acids long and differing only by two conservative substitutions, a valine for an isoleucine in one position and an arginine for a lysine in a second. Our review also showed that the histones of the chromatins of different organs of a given creature are all the same five classes of histones, that is, they are not organ-specific, and, as shown here, they are not species-specific.

"In 1968, with so little known of the chemistry or structure of histones, and with an emerging interest in this subject as a result of the great advances in our knowledge of DNA structure and chemistry, our review found a very great response.

"The members of my group associated with the study of isolated chromatin are all authors of this review, and their names appear in alphabetical order. Luckily, my name begins with 'B' so I'm first. Fambrough is now a staff member of the Carnegie Institution Laboratory of Embryology on the Johns Hopkins campus in Baltimore.

"There have been two reviews of the field of chromosome structure since our 1968 review."<sup>4,5</sup>

1. Huang R C & Bonner J. Histone. a suppressor of chromosomal RNA synthesis. *Proc. Nat. Acad. Sci. US* 48:1216-22, 1962.
2. Huang R C. Citation Classic. Commentary on *Proc. Nat. Acad. Sci. US* 48:1216-22. 1962. *Current Contents* (12):9, 20 March 1978.
3. Allfrey V G, Littau V C & Mirsky A E. On the role of histones in regulating ribonucleic acid synthesis in the cell nucleus. *Proc. Nat. Acad. Sci. US* 49:414-19. 1963. (Cited 545 times.)
4. Kornberg R D. Structure of chromatin. *Annu. Rev. Biochem.* 46:931-54. 1977. (Cited 705 times.)
5. Ts'o P O P, ed. *The molecular biology of the mammalian genetic apparatus*. Amsterdam: North-Holland. 1977. 2 vols.