HISTONES AND HMG-LIKE PROTEINS FROM PISUM SATIVUM CORRELATED WITH EQUIVALENT FRACTIONS OF CALF THYMUS

Herlt, M. Institute of Genetics
University of Bonn, Federal Republic of Germany

Eucaryotic DNA is closely associated with a wide variety of DNA-binding proteins. Traditionally, they are divided into two different classes, the histones and the nonhistone chromosomal proteins. Due to the basic nature of the histones, caused by the high proportion of positively charged amino acids such as lysine and arginine, these proteins bind tightly to DNA nearly all the time and regardless of its nucleotide sequence. Histones act as structural elements for folding the DNA into nucleosomal strands. The fundamental packing unit, the nucleosome, is formed by the core histones H2A, H2B, H3, and H4. H1 histones appear to be responsible for organizing nucleosomes into regular higher-order structures. The heterogeneity in histone H1 as well as the high rate of post-translational modification events suggest a functional role by the induction of conformational changes in the chromatin structure.

The abundant group of nonhistone proteins (NHP's) comprises the remaining DNA-binding proteins. Due to their low concentration in the nucleus, these putative gene regulatory proteins are very difficult to isolate in sufficient amounts. A well defined class of NHP's is represented by the high mobility group or HMG proteins, so called because they are relatively small and highly charged and, therefore, move quickly during electrophoresis. HMG 14 and HMG 17 are characterized by their specific interaction with nucleosomes associated with active genes. At present participation of the HMG-proteins in gene regulation is described only in animals. Investigations aimed at identifying HMG-equivalents in plants are needed.

In this paper Fig. 1 shows an electropherogram of a 10-20% SDS-gel (Laemmli-system) comparing purified histone fractions of calf thymus with Pisum sativum. A difference can be seen in the electrophoretic mobilities of the H2-group proteins (lane 2,4, arrows). This plant-specific protein pattern of H2A/H2B (lane 4) is reconfirmed in comparisons of histone H1 complexes (purified by gel filtration). The two main variants of calf thymus HI (lane 3) exhibit a relative molecular mass of about 30 KD. In contrast, at least three pea HI bands are found at about 40 KD.

Fig. 2 represents an SDS-PAGE of HMG proteins. The polypeptide pattern of a crude HMG protein preparation of calf thymus is shown in lane 2, HMG 1 and HMG 2 enriched by ion-exchange chromatography in lane 3, HMG 14 and HMG 17 purified by preparative electrophoresis in lane 4 and lane 5, respectively. Lane 6 exhibits an equivalent HMG preparation of Pisum. The arrowheads indicate the main HMG polypeptides. Faint bands located at about 40 KD are assumed to be histone HI contaminations.

The similarity in extractability and electrophoretic mobility of mammalian HMG's and the corresponding pea protein fraction permits the conclusion that HMG equivalents exist in plants. However, in the case of HMG's as well as histone H2-group and H1 complex a plant specificity can be supposed.

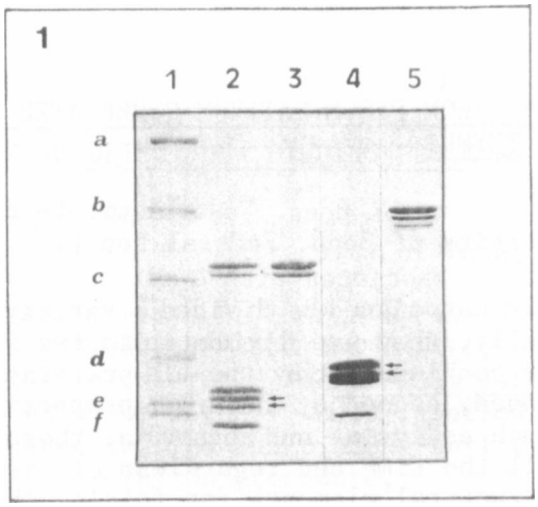

Fig. 1. SDS-Polyacrylamide gel electrophoresis (PAGE) of histone fractions.
Lane 1: Molecular weight standards

a) bovine albumin	66 KD
b) ovalbumin	45 KD
c) carbonic anhydrase	29 KD
d) trypsin inhibitor	20 KD
e) myoglobin	17 KD
f) cytochrome	12 KD

Lane 2: Total calf thymus histones
Lane 3: Histone H1 of calf thymus
Lane 4: Core histone fraction of pea
Lane 5: Histone H1 of pea

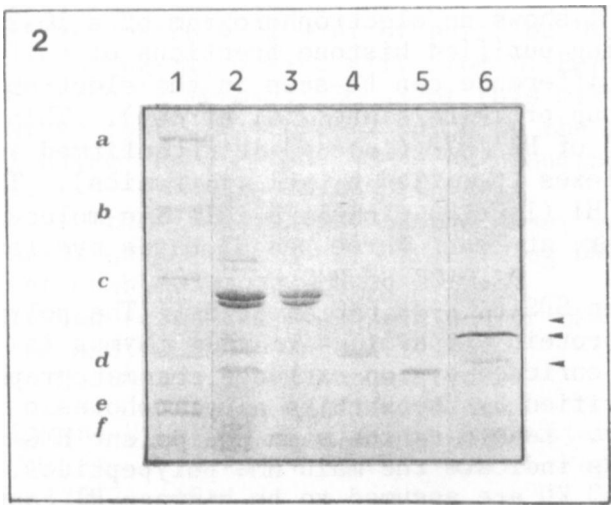

Fig. 2. SDS-Polyacrylamide gele electrophoresis of high mobility group (HMG) proteins:
Lane 1: Molecular weight standards as indicated in Fig. 1
Lane 2-5: HMG fractions of calf thymus
Lane 2: Crude HMG preparation
Lane 3: HMG 1 and HMG 2
Lane 4: HMG 14
Lane 5: HMG 17
Lane 6: HMG preparation of pea

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