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Britten R J & Kohne D E. Repeated sequences in DNA. *Science* **161**:529-40, 1968.
[Department of Terrestrial Magnetism, Carnegie Institution of Washington, DC]

This paper showed the existence of large quantities of repeated sequences in the genomes of higher organisms and demonstrated the use of kinetics of reassociation to determine the "complexity" or amount of different sequences in DNA. [The *SCI*® indicates that this paper has been cited in over 1,905 publications since 1968.]

Roy J. Britten
Kerckhoff Marine Laboratory
California Institute of Technology
Corona del Mar, CA 92625

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My colleagues in the Department of Terrestrial Magnetism developed the agar method[1] for hybridizing DNA. However, their measurements of hybridization between the genomes of higher organisms could not be explained, which led to my proposal of the existence of repeated sequence families. Once the repeats were recognized, the earlier measurements of interspecies relationships showed dynamic processes of evolutionary change of repeats. Also important were studies of thermal stability of hybrids[2] and the demonstration[3] that only a subset of the DNA formed hybrids.

In the spring of 1964, the first tests of the unexpectedly fast hybridization rates showed that agar did not have a catalytic effect on the process. In addition, reassociation of labeled short DNA fragments with large fragments showed that solution reassociation did occur at the fast rates. These early measurements were never published since better methods superseded. However, they were significant because they established that the rate of reassociation of animal DNA was intrinsic in the animal DNA and not an artifact.

In the fall of 1964, Mike Waring and I performed a series of CsCl ultracentrifugation studies on long, denatured mouse DNA. This led to the purification of the mouse satellite DNA and the measurement of its rate of reassociation in solution by optical techniques.[4] The rate of reassociation indicated a repeat length of about 300 nucleotides, which has been fully confirmed.

The intricacies of eukaryotic DNA sequence organization were already indicated in the first year's work. CsCl centrifugation measurements showed that calf DNA and garden pea DNA could be denatured and incubated together to form separate networks yielding two hypersharp bands. We concluded that network formation was DNA sequence-dependent and due to reassociation of interspersed repeats rather than nonspecific aggregation. The DNAs of more closely related species (e.g., calf and salmon) share a few repeated sequences and form single hypersharp bands when incubated together, while they form two bands when incubated separately and mixed in CsCl.

In 1965, Dave Kohne joined our group, and together we carried out the hydroxyapatite studies of reassociation kinetics. The resulting paper is frequently cited because it proved the general occurrence of repeats in eukaryotes, opened a new area of research, and established new techniques and a quantitative approach. Studies of repeats remain very much alive because of their connection to mobile genetic elements. Modern observations show rapid evolutionary frequency changes of interspersed repeat families. It is very likely that many repeats are or were transpositions or were inserted by their action, which has affected DNA sequence organization in nearly every kilobase of the genome of most higher organisms.

1. **McCarthy B J & Bolton E T.** An approach to the measurement of genetic relatedness among organisms. *Proc. Nat. Acad. Sci. US* **50**:156-64, 1963. (Cited 225 times.)
2. **Martin M A & Hoyer B H.** Thermal stabilities and species specificities of reannealed animal deoxyribonucleic acids. *Biochemistry* 5:2706-13, 1966. (Cited 25 times.)
3. **Walker P M B & McLaren A.** Specific duplex formation *in vitro* of mammalian DNA. *J. Mol. Biol.* 12:394-409, 1965. (Cited 65 times.)
4. **Waring M & Britten R J.** Nucleotide sequence repetition: a rapidly reassociating fraction of mouse DNA. *Science* **154**:791-4, 1966. (Cited 190 times.)