COLCHICINE-INDUCED ABNORMAL MEIOTIC CHROMOSOMAL SEGREGATION IN PRIMARY OOCYTES OF THE CHINESE HAMSTER

Part I. NONDISJUNCTION

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Summary In vivo genome mutagenicity of colchicine in the primary oocytes of the Chinese hamster was analyzed at a selected dose that did not completely arrest the formation of the first meiotic spindle, but exhibited a remarkable induction of nondisjunction, very possibly as the result of the inhibition of tubulin polymerization.

A single dose of 3 μg/g b.w. colchicine was intraperitoneally administered to females with a normal estrous cycle at the onset of the formation of the first meiotic spindle. Chromosome analysis of a total of 2,124 secondary oocytes revealed that the incidence of aneuploids increased significantly (p<0.001) in the experimental group (25.9%, 99 out of 382) as compared with the control group (2.0%, 35 out of 1,742). The minimum genome mutagenicity of colchicine caused nondisjunction in one or two bivalents and eventually induced a large number of aneuploid secondary oocytes within haploid range.

The condition of the spindle in oocytes aged by pre- and postovulatory overripeness, both of which have been known to induce meiotic chromosomal nondisjunction, were discussed in relation to tubulin polymerization.

INTRODUCTION

It has been confirmed that colcemid induces aneuploidy in mammalian somatic cells in vitro, owing to its deleterious effect on the mitotic spindle (Cox and Puck, 1969; Kato and Yoshida, 1970, 1971; Cox, 1973). Prior to this, in vivo administration of colchicine has been found to suppress the second meiotic chromosomal segregation of oocytes, thereby inducing triploids in rodents (Edwards, 1954, 1958; Piko and Bomsel-Helmreich, 1960; McGaughey and Chang, 1969). These studies suggested that aneuploids were also produced, but unfortunately, the chromosomal
technique was not sufficiently developed yet to demonstrate with accuracy the genome mutagenicity of colchicine. Today, however, it is possible to undertake a much more informative experiment, since the chromosomal technique for mammalian oocytes has recently been greatly advanced (Kamiguchi et al., 1976, 1978).

Considering the recent achievements in the heteromorphic marker chromosome studies which have shown that nondisjunction seems to occur more frequently in meiosis I than in meiosis II of oocytes (Jacobs and Morton, 1977; Niikawa et al., 1977; Mattei et al., 1979), we attempted to examine the effect of colchicine on meiosis I of oocytes in vivo. The present paper describes the genome mutagenic effect of colchicine at a selected dose that did not completely arrest the formation of the first meiotic spindle of the Chinese hamster's oocytes, but exhibited a remarkable induction of nondisjunction.

MATERIALS AND METHODS

Virgin female Chinese hamsters aged 5–8 months were used. Under a constant laboratory condition, 14 hr illumination from 5:00 to 19:00, temperature at 23±2°C and humidity between 50-60%, they maintained a stable 4-day estrous cycle. The surge of LH occurred at 14:00–15:00 of the day of proestrus and ovulation did at 3:30–4:30 the following morning. A single dose of 3 μg/g b.w. colchicine was intraperitoneally injected to the females at 17:30 of the day of proestrus. We could be certain that the eggs were exposed to the chemical at the onset of spindle formation and thereafter. At about 9:00 of the next day, i.e., of the day of estrum, secondary oocytes were collected from the ampullar region of oviducts.

Chromosomal preparations were made by the method reported previously by us (Kamiguchi et al., 1976, 1978). The technique has been elaborated so as to avoid rupturing the plasma membrane of oocytes, in order to minimize the rate of accidental loss of chromosomes. Using this method, chromosomes of oocytes can be properly spread within flattened dry ooplasm without being scattered. Thus, more than 90% of the secondary oocytes were karyotyped successfully. In the Chinese hamster, metaphase II consists of eleven dyads which can be divided into four morphologically distinguishable groups, i.e., two large metacentric (Group A), three medium metacentric (Group B), three medium acrocentric (Group C) and three small metacentric chromosomes (Group D). Aberrant dyads were identified according to the karyologic system.

RESULTS

The incidence of aneuploid oocytes in the experimental group was much higher (p<0.001) than in the control group (Table 1). Thus, the induction of nondisjunction by colchicine in the first meiotic division of oocytes was unequivocally