2h and 4h the labelling of rRNA and its precursors is greatly increased.

By using 7.5% gels and isolating the 1.1A 2 and 4h after oestradiol treatment, large increases are observed in labelling of an RNA peak associated with the 4S RNA.

Actinomycin D affects synthesis of the various RNA species differentially. rRNA species are most affected and a dose of 30 µg/rat completely eliminates increased labelling of these fractions in response to oestradiol. The RNA that remains at the origin and tRNA of the gel are least affected and small extents of incorporation are observed even at actinomycin D doses of 125 µg/rat.

Cycloheximide at a dose causing 98% inhibition of protein synthesis, given before or simultaneously with oestradiol, seriously decreases the incorporation of precursors into all species of RNA. When administered 1h after the hormone its greatest effect is on the maturation of rRNA.

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Alkylated Components in Rat Liver Ribonucleic Acid: a Possible Index of True Biological Turnover

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In comparative studies of alkylating agents of different carcinogenic potency methyl methanesulphonate was injected into rats at a dose of 50mg/kg. Earlier observations had indicated that doses of this order did not produce toxic effects in rats other than a transient effect on late spermatogenesis (Jackson, Fox & Craig, 1961). Recently the time-course of methylation of rat liver RNA *in vivo* was followed for the first 24h after a similar dose (Whittle, 1969). Methylation was shown to be maximal by 2h and apparently remained constant until 24h, in contrast with the reported loss of 40% of methylated components by 24h when the carcinogen dimethylnitrosamine was used (Craddock & Magee, 1963). The study with methyl methanesulphonate was extended over a 14-day period, during which the apparent half-life for the biological turnover of rRNA was determined after a single injection of [5-³H]orotic acid and compared with the rate of disappearance of 7-alkylguanine

from RNA. To avoid any loss of RNA (Whittle & Potter, 1968) food intake was not restricted.

Two procedures were employed for the purification of RNA: the method of Ralph & Bellamy (1964), which preferentially extracts rRNA and tRNA, and Kirby's (1965) procedure, which extracts rRNA and some rapidly labelled RNA. The apparent half-life of rRNA determined by using [³H]orotic acid was approx. 5.0 days in both the control series and the methyl methanesulphonate-treated rats, a value in agreement with other reports (Blobel & Potter, 1968; Avdalović, 1970), indicating that the treatment had no detectable effect on the rate of breakdown of rRNA. The loss of 7-methylguanine was followed by analysis of acid hydrolysates of rat liver RNA preparations by using Dowex 50 (H⁺ form) column chromatography. The maximum extent of alkylation was 0.040–0.046% guanine and the loss of 7-methylguanine followed first-order kinetics with a half-life of approx. 3.5 days. The half-life of rRNA determined by orotic acid labelling must be an overestimate, owing to the reincorporation of labelled RNA degradation products. Since it is known that the glycosidic link of 7-methylguanosine is chemically stable at neutral pH (Kriek & Emmelot, 1964) the evidence strongly suggests that loss of 7-methylguanine under these conditions may give a better index of the true turnover of rat liver RNA.

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Studies on the Methylated Regions of HeLa-Cell Ribosomal and Nucleolar Ribonucleic Acids

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The methylation patterns of HeLa-cell rRNA and nucleolar RNA have been examined by the 'finger-printing' technique of Sanger, Brownlee & Barrell (1965) with RNA preparations methyl-labelled