Danieli, G. A. and E. Rodinò. University of Padua, Italy. Biochemical estimate of DNA content in D. hydei salivary glands.

Practically all the investigations on nucleic acids content in salivary glands are carried out by means of cytochemical and histophotometric methods. Actually the biochemical approach to the problem is quite infrequent in literature. The average content of DNA and RNA per cell was determined by Patterson and Dackerman (1952) in D. melanogaster salivary glands by chemical methods. The determinations were made on 18 pairs of glands by a microtechnique (Linderstrøm-Lang and Holter apparatus) using a modification of the Schneider method.

We have decided to investigate the same problem using the improved Burton's DPA method. In recent years this method was applied successfully on various materials for evaluations of extreme sensitivity coupled with low reagent blanck. In order to obtain a further increase in sensitivity, we followed the Giles and Myers (1965) suggestions. Preliminary experiments pointed out that the best results are obtained with a 2.5% DPA concentration. Drosophila hydei salivary glands have been studied at various larval stages from the late second to the late third instar.

From synchronized cultures, 100 larvae per point were dissected in Ephrussi-Ringer solution at 4°C temperature in order to reduce metabolic activity. The glands were collected and immediately frozen on dry ice, and then stored at -30°C until used.

Extractions and determinations were carried out at the same time on all the groups of glands.

After thawing, the glands were homogenized in 10% TCA and the material was subsequently treated according to the standard Schneider procedure with centrifugation at 4°C, 4,000 r.p.m. for 30 minutes in 5% TCA followed by treatment with ethanol and ether.

The nucleic acids were extracted from the dried residue with 1.6 ml. of 5% TCA at 90°C for for 15 minutes, the material extracted was then centrifuged at 4,000 r.p.m. for 45 minutes.

1 ml. of supernatant was used for DNA determination. The colorimetric reaction was that of Burton's modified DPA method.

Some of our first results are reported in Table 1.

Table 1

age of larvae (hrs. after ovoposition)	average DNA content per gland expressed in ug. x 10^{-2}
96	2.85
144	8.12
192	12.02

A comparison between the absolute values of DNA content per cell found by several authors in D. melanogaster and in D. hydei is given in Table 2.

Table 2

Patterson and Dackerman 1952 (biochemical evaluation)	D. melanogaster	2.8
Kurnick and Herskowitz 1952 (histophotometric evaluation)	D. melanogaster	0.712
Plaut 1963 (quantitative autoradiography)	D. melanogaster	about 10
Berendes 1965 (histophotometric evaluation)	D. hydei	7.05 (distal nuclei)
Our results (biochemical evaluation)	D. hydei	9.2

The values are expressed in ug. x 10^{-4}

References: Berendes, H. D. 1965. Chromosoma, 17:35; Burton, K. 1956. Biochem. J., 62:315; Giles, K. W. and Myers, A. 1965. Nature, 206:93; Kurnick, N. B. and Herskowitz, I. H. 1952 J. Cell. Comp. Physiol. 39:281; Patterson, E. K. and Dackerman, M. E. 1952. Arch. Biochem. and Biophys., 36:97; Plaut, W. 1963. J. Mol. Biol. 7:632.