On the other hand, the mean value of the five rats receiving thiouracil (rats 62–66) shows a clear tendency to increase.

In order to demonstrate more clearly these variations, we pooled all the measurements in each group; Rat 61 was not included, since its mean value was not 'consistent' with the four other rats of the same group (Table 2).

Table 2. POOLED RESULTS

	n	$\bar{x}$	$S\bar{x}$	S
Controls	443	834	± 8·6	179·7
Thyroxine	383	737	± 9·4	185·0
Thiouracil	480	879	± 8·4	185·8

By 'Student's' t-test we have demonstrated that the observed variations are highly significant (Table 3).

Table 3. t-TEST

	t	P
Controls/thyroxine	7·5	< 0·001
Controls/thiouracil	3·7	< 0·001
Thyroxine/thiouracil	11·0	< 0·001

By these experiments we have demonstrated the existence, in the thyroid of the white rat, of a correlation between the variations in cell activity and the deoxyribonucleic acid content of the nuclei; the deoxyribonucleic acid increases with stimulation of cell activity and decreases with inhibition of cell activity. The deoxyribonucleic acid content of the thyroid of normal rats, and especially those with thyroxine-inhibited thyroids, is markedly below the diploid value; on the other hand, in the stimulated thyroid, the content of deoxyribonucleic acid reaches, approximately, the theoretical diploid value.

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Composition and Action of Yeast Polygalacturonase

YEAST polygalacturonase is an exo-cellular pectic enzyme produced by *Saccharomyces fragilis* in a synthetic, protein-free medium. The enzyme is constitutive and is not accompanied by pectin-esterase[1]. When it is allowed to act on pectic acid, the following series of reactions occurs[2]:

(*a*) Pectic acid → penta- + tetra- + tri- + digalacturonic acids;

(*b*) Pentagalacturonic acid → tetra- + galacturonic acids;
Pentagalacturonic acid → tri- + digalacturonic acids;

(*c*) Tetragalacturonic acid → tri- + galacturonic acids;

(*d*) Trigalacturonic acid → di- + galacturonic acids.

Reactions (*a*) and (*b*) comprise the initial rapid linear phase of the hydrolysis. The optimum pH of (*a*) is 4·4. A slower linear phase, beginning at 25 per cent hydrolysis, is characterized by reaction (*c*) which is optimal at a pH of approximately 3·5. After about 50 per cent hydrolysis, (*d*) is the main reaction, and it occurs at a very low rate. Finally, after 70 per cent hydrolysis, the reaction ceases, leaving the dimer and monomer as end products. Reaction (*d*), like (*c*), is much slower at the higher pH; hence at pH 4·5, the reaction appears to cease at 50 per cent hydrolysis and the apparent end products are tri-, di- and galacturonic acids[3].

In view of the postulations of Dingle *et al.*[4] and Schubert[5] which suggest that several polygalacturonase components occur in mould pectic enzymes, the nature of yeast polygalacturonase was studied.

We have found that yeast polygalacturonase of the crude culture liquid can be adsorbed in the cold on a pectic acid gel at pH 3·0 and eluted with 1 N acetate buffer at pH 5·0. This resulted in a 13-fold concentration, but only in a negligible increase in specific activity. An electrophoretic analysis was performed after further concentration by precipitation with saturated ammonium sulphate followed by dialysis. The pattern indicated two components. The major one comprised about 95 per cent of the total protein. A variable solvent solubility test also indicated two fractions. At 4° C., the minor fraction was precipitated at 0·2 ammonium sulphate saturation and the major one was salted out between 0·8 and complete saturation. A specific property solubility test based on the procedure of Falconer and Taylor[6] showed that only the major fraction had yeast polygalacturonase activity. The specific activity of this fraction, calculated by the last method, was 0·179 yeast polygalacturonase unit per mgm. protein (1 unit of activity releases 1 mM of reducing groups per min. at pH 5·0 from a 0·5 per cent pectic acid solution). The material which was precipitated between 0·75 and complete saturation moved as a single boundary in the ultra-centrifuge.

The rates of yeast polygalacturonase activity on tri-, tetra- and poly-galacturonic acids were compared, using the filtrates obtained at four different concentrations of ammonium sulphate. The ratios of activity remained roughly constant during the salting out, indicating that the hydrolysis of pectic acid to the digalacturonic acid stage is catalysed by a single enzyme. Full details of this work will be published elsewhere.

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