STUDIES ON THE EFFECT OF SOME PESTICIDES ON THE CHEMICAL COMPOSITION OF SOME YEASTS

A THESIS

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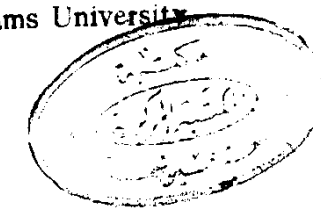

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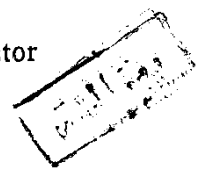

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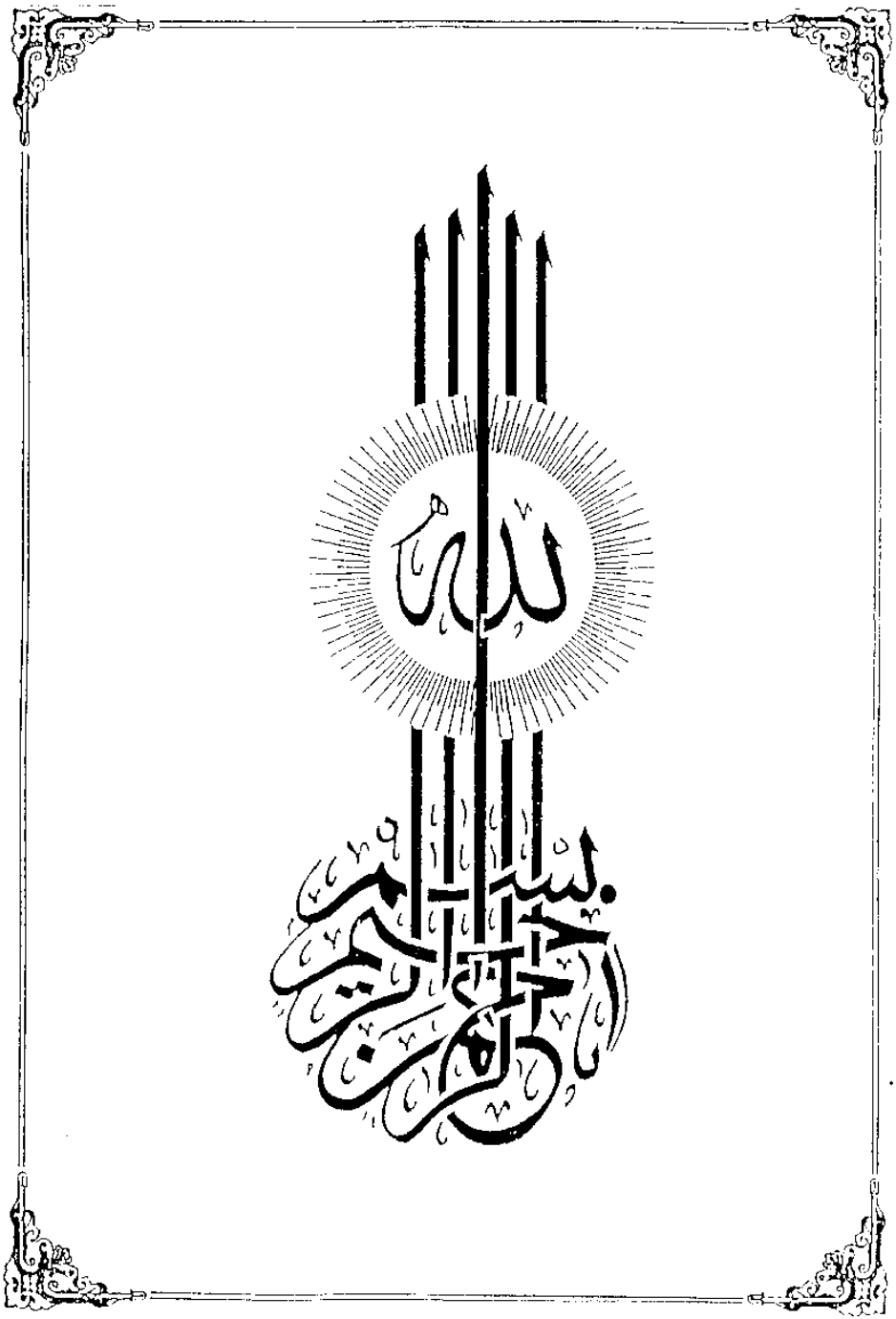

"THANKS TO OUR MIGHTY GOD FOR HIS

CONTINUOUS SUPPORT TO ME "

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INTRODUCTION

Yeasts are widely distributed in nature. They thrive on various parts of plants, like the surface of both leaves and fruits. Also the soil forms an important reservoir for yeasts, where they can survive unfavorable periods. The persistence of pesticide residues in the invironment is marked, and can interfere with the growth and activity of microorganisms which are largely responsible for continued soil fertility. Therefore the inhibitory effect of pesticide programs on the non-target soil and epiphytic microorganisms has recently attracted the attension of investigators.

The present study, however, is a trial to demonstrate and investigate the changes in the levels of different fractions of cellular carbohydrates, lipids, proteins and free amino acids of three yeasts originally isolated from the surface of some fruits, as they were grown in the presence of different sublethal concentrations of three commonly used pesticides namely; the fungicide (Tilt), the insecticide (Dimicron) and the acaricide (Neoron). The organisms under investigations are the ascosporagenous yeast Hansenula anomala and other two anascosporagenous yeasts namely the pigmented Rhodotorula rubra and the non-pigmented Candida guilliermondii.

I- REVIEW OF LITERATURE

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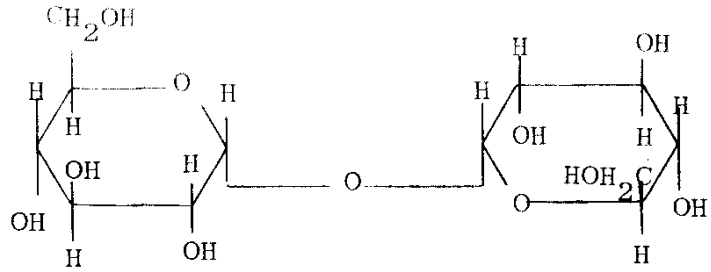
I.1. Yeast carbohydrates:-

Yeast carbohydrates can be divided into two main categories. These are the storage and structural carbohydrates.The storage reserve carbohydrates in yeasts are trehalose and glycogen, while the structural carbohydrates are mostly mannan and glucan.

Chester (1963) found that about 40% of the dry weight of anaerobically grown yeast cells may be reserve carbohydrates. Also, Travelyan (1958) reported that the cell wall of Saccharomyces cerevisiae contains B-glucan and mannan.

Trehalose was first isolated from pressed Baker's yeast by Koch and Koch (1925). It was crystallized from methanol extracts and was identified on the basis of melting point, optical rotation. Travelyan and Harrison (1952) have used trichloroactic acid to extract trehalose from yeast cells.

The chemical structure of trehalose is (α-D-glucopyranosyl- α-D-glucopyranoside).



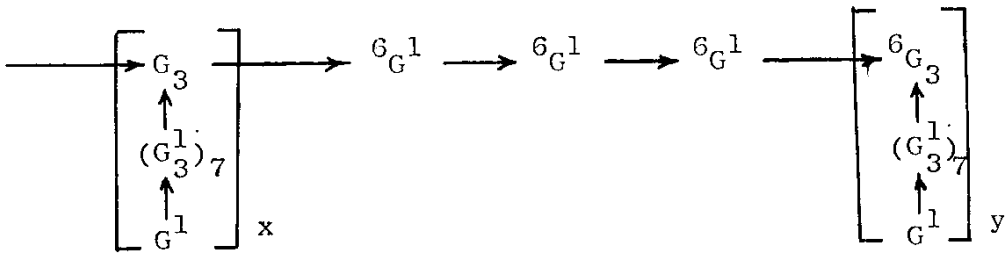
It is a non-reducing disaccharide composed of two glucose residues.

Glycogen whether from animal or microbiol cells are now generally accepted to be multi branched molecules of high molecular weight which consists of numerous chains of α-(1→4) lin ed D-glucose residues. These chains normally contain an average of about 12-D-glucose residues, although individual chains vary considerably in length, and are arranged to form a tree or bush-like structure (Manners, 1957).

Yeast mannan, which at the time of discovery by Salkowski (1894) was termed "yeast gum" was first studied in detail by Haworth et al., (1937, 1941). It could be extracted from whole yeast by boiling with 6% NaOH and precipitated as its insoluble cupper complex by addition of Fehling's solution to the alkaline extract.

Salkowski (1894) prepared from yeast an insoluble polysaccharide which was termed "yeast cellulose" which was then known as glucan. Zechmeister and Toth (1934, 1936) showed that it contained a preponderauce of (1-3) linkages. Barry and Dillon (1943) established the final structure of yeast glucan as B-(1-3) linked D-glucose residues together with little B-(1-6) residues.

Yeast glucan was isolated after drastic treatment of whole yeast first with hot dilute alkali followed by heating for several hours with dilute HCl.



$x + y = 40\text{-}50$

Schematic representation of Baker's yeast glucan according to Misaki et al., (1968).

Elinova and Vitovskaya, 1963; Takumi and Kenichi, 1968; Elinova and Vitovskaya, 1971; Elinova and Vitovskaya, 1972; Kaluskin, 1974; Katohda, et al.,1976 and Slodki, 1980 studied the different fraction of polysaccharides

in pigmented and non-pigmented yeats. They pointed out that non-pigmented organisms contained mostly mannose and small amounts of glucose, while the red species consisted mainly of mannose (23.2-71 %) with minor amounts of glucose and galactose, and in some others very small amounts of both xylose and fucose but in yellow species except Rhodotorula flava, their polysaccharides contained mannose (24-36.2%), xylose (14-23%), glucose (13.5-44%) as well as a uronic acid component.

Tokuyo et al. 1963 studied the composition of extracellular polysaccharides of the subgenus Flavotorula and Rubrotorula and indicated that all Flavotorula strains produced extracellular amylose which stained blue with Iodine, whereas the polysaccharide produced by Rubrotorula strain gave no blue color reaction with Iodine.

The solution of polysaccharide produced by the Flavotorula strains were more viscous than those of the Rubrotorula strains. The polysaccharide produced by the Flavotorula were principally composed of xylose, mannose, glucose and galactose whereas galactose and xylose were not found in the Rubrotorula strains. The study was contined by Elinova and Vitosskaya, 1979 who indicated that the mannan produced by Rhodotorula

rubra strain has alternating B- (1→3) and B-(1→4) linkages.

Several investigations have been published on the chemical composition and properties of polysaccharides isolated from different species of Candida (Elinova and Drobyazgo, 1965; Masler et al., 1966; Surinova,1968; Strychkova et al., 1971). Petrushko and Kalyuzhnyi, 1971 working on Candida species, also determined the lyophilic and lyophobic properties of the cell wall, and noticed that the ratio of glucomannan to glucan polysaccharides had some effect on the flotation ability of Candida, and its concentration ranged between 0.47-1.24%. A more direct effect on the flotation qualities was also obsereved in the chitin content, where high chitin content improved flotation.

The main carbohydrates of yeast cells,other tham the structural polysaccharide glucan and mannan are glycogen and trehalose (Trevelyan, 1958). These compounds occur in high proportion, if the yeast was grown under suitable conditions. Mannan and glucan are synthesized in parallel, as would be expected since both are constituents of the cell wall. Their formation is not reduced by nitrogen assimilation, as in the formation of trehalose and glycogen (Trevelyan and Harrison, 1956).

Glycogen, unlike mannan or glucan is not confined to the cell wall (Northcote, 1953), and its occurence in different regions of the cell means that glycogen is not uniformly free to diffuse out of the cell when yeast is treated with reagents such as carbonate or acetic acid.

Northcote and Horne, 1952; and Roelofson, 1953 found that glucan was present in yeast cells up to 29%. Also, glucan and mannan were synthesized by different strains of Saccharomyces species grown on glucose containing media, and their synthesis proceeded via the formation of glucose and mannose phosphates (Chung and Nickerson, 1954).

Filippova, (1982) noticed that the polysaccharides isolated from Rhodotorula gracilis, Sporobolomyces pararoseus, Candida tropicalis, and Candida scottii constituted 25-30 % of the dry biomass and contained trehalose, glycogen and glucan. The chemical composition of the polysaccharide was not significantly changed when the yeasts were cultivated in different culture media. He also noticed that chitin constituted 0.11-0.30% of the cell wall.