TABLE OF CONTENTS

			Page	
ı.	Introduc	tion .	1	
II.	Review o	f literature	3	
	2.1	Pectic substances	3	
		Definition and structure Distribution of pectic substances in	3	
	2.1.3	plants Properties and application of pectic	5	
	2.1.5	substances.	6	
	2.2	Pectic enzymes	8	
	2.2.1	Nomenclature and classification	8	
	2.2.2	Pectinesterases	9	
	2.2.3	Polygalacturonase	10	
	2.2.4	Pectate lyases	11	
	2.2.5	Pectinlyases	12	
	2.2.6	Other pectic enzymes	13	
	2.3	Microbial pectinases	15	
	2.3.1	Bacterial pectinase	15	
	2.3.2	Fungal pectinases production and		
		properties	18	
	2.4	Use of pectic enzymes in fruit processing		
		technology.	29	
	2.4.1	Fruit juice clarification	29	
	2.4.2	Maceration and liquefication	30	
	2.4.3	Utilization of sugar-beet by-products		
		in production of microbial proteins	33	
	2.5	Protein quality evaluation with special		
		emphasis on fungal proteins	37	

III.	Materia	ls and methods	46
	3.1	Materials	46
	3.1.1	Microorganism	46
	3.1.2	Media used	47
	3.1.3	Pectic substances	49
	3.1.4	Sugar beet by products	49
	3.2	Methods	50
	3.2.1	Determination	50
		3.2.1.1 Determination of polygalacturonase	
		activity	50
		3.2.1.la viscosity reduction method	50
		3.2.1.1b by measuring the increment	
		of reducing groups	51
		3.2.1.2 Determination of pectin lyase	
		(PL) activity	53
		3.2.1.3 Determination of extracellular	
		protein	53
		3.2.1.4 Determination of reducing sugars	54
		3.2.1.5 Determination of total nitrogen	54
		3.2.1.6 Amino acid analysis	54
	3.2.2	Screaning the fungal cultures	55
	3.2.3	Characterization of polygalacturonase	
		type of M. verrucaria .	55
	3.2.4	Factors affecting the physiology of PG	
		production.	56
		3.2.4.1 Effect of incubation period	57
		3.2.4.2 Effect of aeration and agitation	58
		3.2.4.3 Effect of initial pH of cullture	
		medium.	58
		3.2.4.4 Effect of carbon source	58
		3.2.4.5 Effect of beet pulp concentration	59
		3.2.4.6 Effect of nitrogen source.	59
		3.2.4.7 Effect of inoculum size	60

	3.2.5	Biochemi	cal properties of PG activity	60
		3.2.5.1	Effect of enzyme concentration	60
		3.2.5.2	Reaction progress with time	61
		3.2.5.3	Effect of incubation temprature	
		7	on the reaction rate of the	
			enzyme	61
		3.2.5.4	Heat stability of PG activity	61
		3.2.5.5	Effect of substrate concentration	62
		3.2.5.6	Effect of pH on PG activity	62
	3.2.6	Purifica	ation of pectic enzyme	62
		3.2.6.1	Precipitation with acetone	63
		3.2.6.2	Gel filtration of the enzyme on	
			sephadex G 100 coulmn	64
		3.2.6.3	Gel electrophoresis (PAGE)	64
	3.2.7	Animal :	feeding experiment	66
		3.2.7.1	Evaluation of protein quality	
			of fungal proteins of M .	
			verrucaria	69
		3.2.7.2	Calculation of Net protein	
			utilization and utilizable	
			protein (up)	74
		3.2.7.3	Determination of protein	
			efficiency ratio (PER)	74
IV	Results	and Disc	ussion	75
	4.1	Screani	ng of strains of M. verrucaria	
		with re	spect to formation of pectinases	75
	4.2	Screeni	ng the active pectolytic strains	
		of <u>M.</u> <u>v</u>	errucaria with respect to forma-	
		tion of	polygalacturonase (PG) and pectin	
		lyase (PL)	83
	4.3	Charact	erization of polygalacturonase	
		type of	M. verrucaria	85

4.4	Physiology of polygalacturonase formation by M. verrucaria	89
4.4.1	Effect of incubation period on the levels of pectolytic activities of $\underline{\text{M.}}$ verrucaria	89
4.4.2	Levels of endo-polygalacturonase activity produced under conditions of shake and static cultures.	95
4.4.3	Levels of endo-polygalacturonase activity produced under different pH values of the medium.	99
4.4.4	Effect of carbon source on endo-PG formation	102
4.4.5	Effect of beet pulp concentration on levels of endo-PG activity.	107
4.4.6	Effect of inoculum size on endo-PG activity.	109
4.4.7	Effect of nitrogen source on production of endo-PG enzyme	110
4.5	Kinetics and biochemical properties of polygalacturonase activities of M. verrucaria CBS 28846 and CBS 17627	112
4.5.1	Effect of reaction temprature on the reaction rate of crude polygalact-uronase	112
4.5.2	Effect of temprature on stability of crude endo-polygalacturonase	115
4.5.3	Effect of enzyme concentration on endo-polygalacturonase activity.	117
4.5.4	Reaction progress with time	117

4.5.5	Effect of substrate concentration on the	
	reaction rate	120
4.5.6	Effect of pH on crude endo-PG activity	125
4.6	Partial purification of endo-polygalact-	
	uronase(endo-PG)produced by M. verrucaria	128
4.6.1	Fractional preceipitation with acetone	128
4.6.2	Gel filtration of endo-PG on sephadex G 100 column	132
4.6.3	Gel electrophoresis of the partially purified enzyme	138
4.7	Kinetics and biochemical properties of the partially purified enzyme.	140
4.7.1	Effect of reaction temprature on the	
	reaction rate of partially purified	
	enzyme.	140
4.7.2	Effect of temprature on stability of partially purified enzyme	143
	,	113
4.7.3	Effect of enzyme concentration on the reaction rate.	146
4.7.4	Reaction progress with time	149
4.7.5	Effect of substrate concentration on the reaction rate of partially purified enzyme	152
4.7.6		155
4.8	Evaluation of fungal protein from \underline{M} .	
	verrucaria CBS 28846	1 = 0
4.8.1	Chemical evaluation of gungal protein	158

	4.8.2	Animal	feeding	experiment	163
٧.	Summary				170
VT.	Reffere	nces	2		179