

RESULTS AND DISCUSSION

I. Propolis:

A. Propolis Gathering Activity:

The present study was carried out in an apiary at Al-Amar village. This village, Kalubia Governorate is located about 40 km far from Cairo. The apiary consisted of 100 colonies. The apiary was surrounded by cultivated land, planted with many different plants throughout the year. The most important sources of nectar and pollen in spring were apricot and citrus which begin their blooming season from the first days of February to the end of the same month for apricot, then the blooming of citrus begins in the second half of March to the second two weeks of April. The third source is clover which spread at Kaha where the apiary was located. The blooming of clover begins at May and extend to the middle of June, while the fourth source of nectar is cotton. Meanwhile, the main source of pollen is maize. Both continue blooming till September. Beside these main sources there are some minor sources throughout the year such as sunflower, and vegetables.

Bees collect propolis from the resins and secretions of buds from the bark of resinous and deciduous trees around the apiary. Survey on sources of propolis revealed that trees are the main sources. These plant sources are responsible for the wide variations in propolis physical and

chemical properties. The trees which was found to be as the main sources of propolis at Al-Amar, and Kaha areas were Acacia farnesiana, A. nilotica, Populus nigra, Eucalyptus comaldulensis, Robinia pseudoacacia, Capiessus semp, Prunus persica, Prunus domestica, Prunus armeniaca, Pyrus communis, and Salix aegyptiaca. So colour of Egyptian propolis is yellowish green, yellowish-brown, dark-brown, and black greenish (Fig. 2) Propolis colour is varied according to the source of pollen. The smell of Egyptian propolis is similar in general. The odour is obvious, when we keep propolis in close size.

The pollen grains of the above trees were found in the insoluble fraction of the propolis gathered.

These results are in agreement with Ioyrish (1974) who stated that it used to be thought that propolis was made from substances collected by bees from the buds of trees, but research has established that it is prepared from pollen.

Caillas (1978) mentioned that propolis might be a resin residue coming from the first phase of pollen digestion in small organ placed between the sac and the lower gut.

Konig (1985) stated that the geographical range of popular is limited to N. America, Europe, N. Africa and the non-tropical regions. In Africa, A. mellifera has a more extensive range than that of poplar and is found through the whole continent.

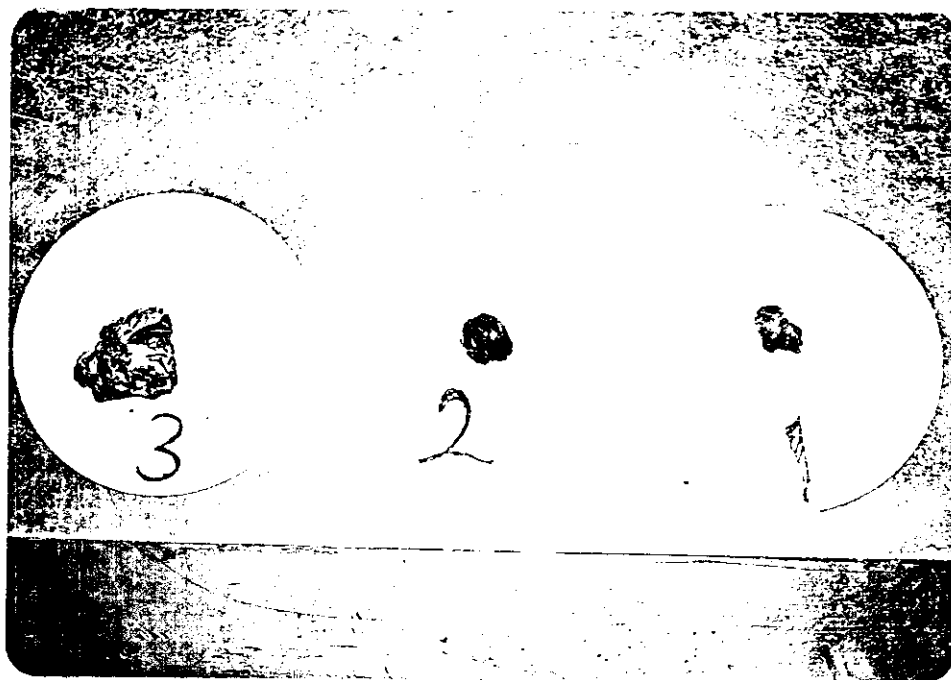


Fig. (2): Crude propolis colour (1 and 3) and (2) pitch material collected by honeybees.

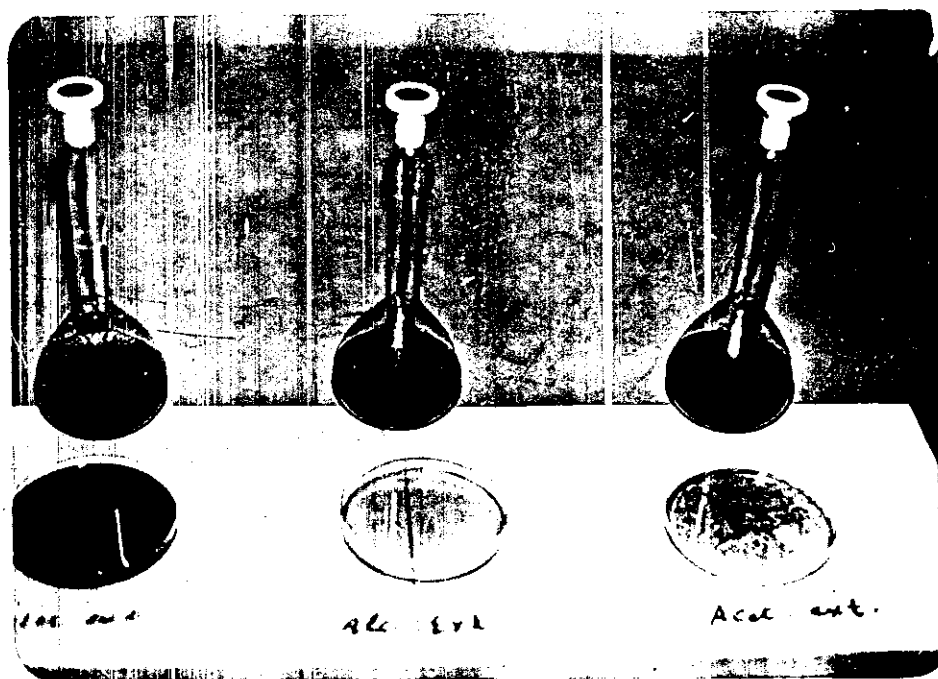


Fig. (3): Propolis extracts colour, 1 water ext., 2 ethanol ext. and 3 acetone ext.

Omar (1988) in upper Egypt, found that propolis colour varies from yellow-green to dark brown in different localities and months.

Propolis substitutes:

We noticed that on February, in time of need, bees collected resinous substitute materials such as pitch (Fig.2). This result is in agreement with Taber and Barker (1974), Root (1975), Jaycox (1980) and Lowe (1980). They recorded that where sources of propolis appear absent bees use propolis substitutes such as chaulking compound, pitch and certain man-made products, like paint, bitumen or mineral oils. The increase of such substitutes depends on the decrease of the natural propolis sources.

B. Collection of propolis:

Propolis gathers are usually the older bees who brake off pieces of resinous, exudate from tree buds or branches, by using their mouth-parts and hind legs. The pieces are moistened with the tongue, formed into pellets by the mandibles and passed along the inside of their legs into the corbiculae (pollen baskets). So, the yield of propolis is mainly dependable on species of tree, race of honeybee, season, colony strength and condition of the hive. Propolis which was gathered by each colony of the two races F_1 Italian bees and F_1 Carniolan bees during the period of the experiment had been recorded in Table (1). The data of propolis collection by the two races during a year of study show highly significant differences between the two races and between months (Table 2).

Table (1) shows that F_1 Italian bees gathered 92.95, 61.15, 63.65 and 71.40 g propolis during the year, while F_1 Carniolan bees gathered 45.95, 52.65, 44.25 and 43.55 g.

Meanwhile, monthly amounts of propolis gathered by the two races, F_1 Italian bees from January to December were 2.59, 3.16, 4.31, 7.06, 6.20, 6.60, 10.71, 16.33, 2.93, 4.41, 5.05 and 1.94 g, respectively. As well as, F_1 Carniolan bees in the same period were 1.88, 2.36, 2.75, 4.93, 4.89, 5.5.40, 6.77, 7.40, 2.90, 3.08, 2.45 and 1.80 g, respectively.

The collection of propolis is initiated by some inherent change in the behaviour of foragers and climatic conditions. It is conceivable that, these factors may be interrelated with those which are involved in the use of propolis by bees. So, the great amount of propolis ^{were} was gathered through July and August by both races, Italian and Carniolan were (10.71 g & 16.3 g) and (6.77 g & 7.40 g), respectively (Table 1). While the lowest amount of propolis ^{in Carn} gathered through December and January by F_1 Italian bees, and F_1 Carniolan bees were (1.94 g & 2.59 g) and (1.80 g & 1.88 g), respectively. (Fig. 4).

From the above results, we can conclude that F_1 Italian bees collected more propolis than F_1 Carniolan bees. Also, the rate of propolis collection were more in summer months than in spring and winter months by the two races. These results were in agreement with Mizis (1978) who found that each colony irrespective of race and method of collection can produce from 50 to 700 g propolis, and that with the Caucasian Gray bees, 2-3 times more propolis was obtained. Also, Nilolaev ^k (1978) — mentioned that in one season, the honeybee colony may obtain 100 to 150 g.

Table(1) : The amount of propolis produced monthly during a year of study (from January to December: 1986 . (in grams per colony)

Months	F ₁ Italian bees				F ₁ Carniolan bees				Mean
	1	2	3	4	1	2	3	4	
Col.No.									
January	2.45	2.20	2.65	3.05	2.00	2.70	1.00	1.80	1.88
February	3.55	2.75	2.50	3.85	2.70	2.25	2.00	2.50	2.36
March	4.50	3.95	4.80	4.00	2.50	2.60	2.75	3.15	2.75
April	6.35	7.35	5.05	9.50	4.95	6.70	3.50	4.55	4.93
May	7.40	5.00	5.30	7.10	4.85	3.60	5.70	5.40	4.89
June	8.15	6.45	5.20	6.60	5.35	5.20	4.80	6.25	5.40
July	11.50	8.15	12.70	10.50	6.25	8.15	6.70	6.00	6.77
August.	32.90	10.65	9.50	12.25	5.75	10.50	8.10	5.25	7.40
September	4.50	3.15	3.80	4.25	3.50	3.15	2.80	2.15	2.90
October	4.00	5.20	4.35	4.10	3.60	3.30	2.40	3.00	3.03
November	5.50	4.40	6.30	4.00	2.50	3.10	2.70	1.50	2.45
December	2.15	1.90	1.50	2.20	2.00	1.40	1.80	2.00	1.80
Total	92.95	61.15	63.65	71.40	45.95	52.65	44.25	43.55	46.60

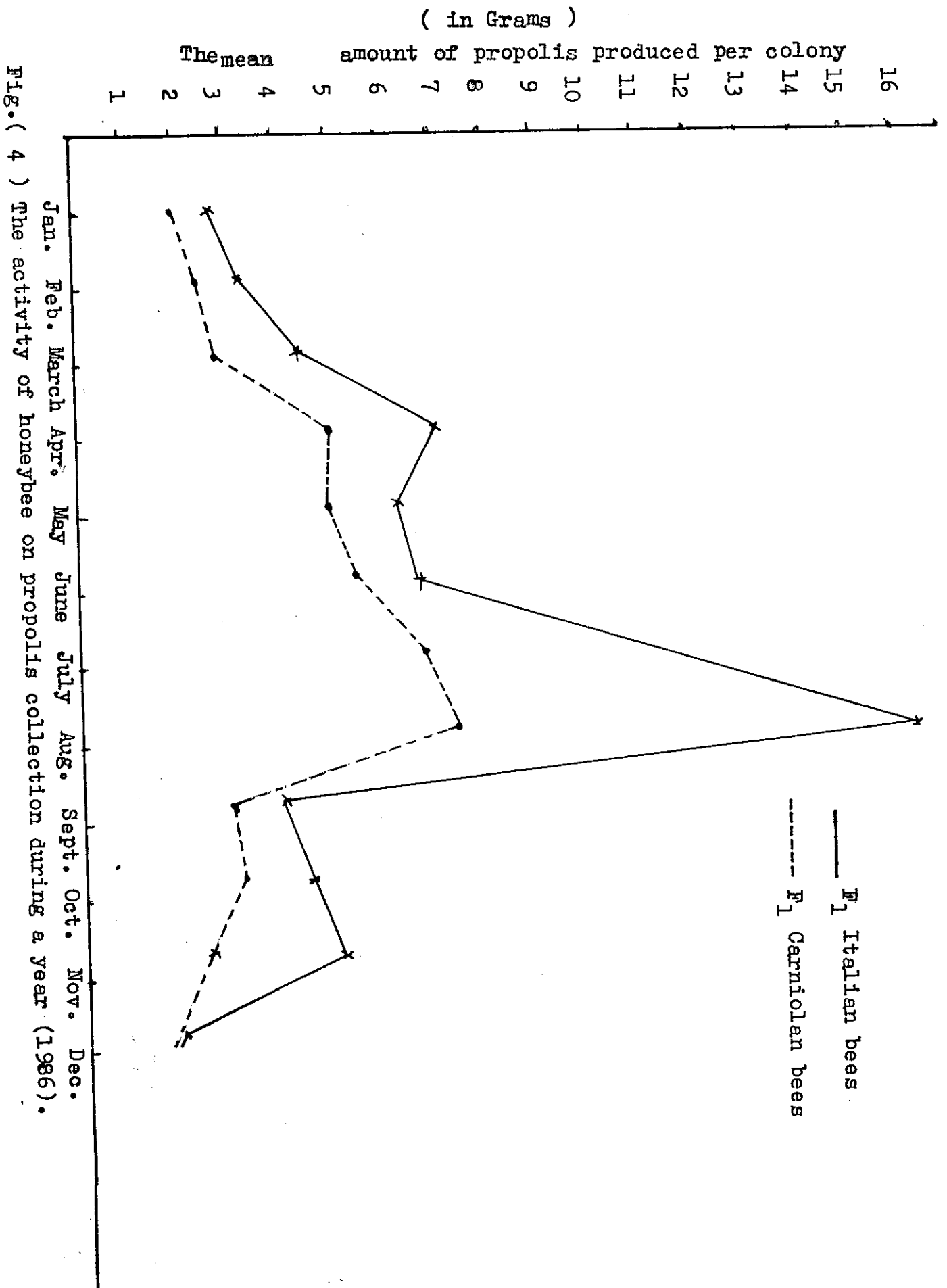
Table (2): Analysis variance of propolis produced during
a year of study.

Source of variance	d.f.	S.S.	M.S.	F.
Colonies	3	167.60		
Breeds (Races of bees)	1	111.47	111.47	6.384**
Months (Dates)	11	751.63	68.33	3.384**
Error	80	1397.26	17.46	

Total	95	1427.93		

* Significant.

** Highly significant.



C. Effect of honeybee races on propolis gathering activity:

In the present study, the total amount of propolis gathered were by Italian bees was 72.29 g/colony per year, while the total amount of propolis were gathered by Carniolan bees was 46.60 g/colony through the year (Table 3). These results show that, F_1 Italian bees collect propolis more activity than F_1 Carniolan bees, which means that F_1 Italian bees gather propolis 55.13% more than F_1 Carniolan bees. Table (3) shows that, the amounts of propolis gathered by F_1 Italian bees colonies were 92.95 g, 61.15 g, 63.65 g and 71.40 g, while the amount of propolis gathered by each colony of F_1 Carniolan bees were 45.95 g, 52.65 g, 44.25 g and 43.55 g.

This result shows that F_1 Italian bees collected more propolis than F_1 Carniolan bees, and that is in agreement with Starostensko (1968) and Krupicka (1972) who reported that some races of honey bees collect propolis more activity than others. Gray mountain Caucasian bee collect rather more than dark forest bees, whereas, Ukranian and Far East bees collect very little. While *Apis mellifera Carnica* bees produces less propolis than native bee under the condition in Czechoslovakia, A.m. Caucasian produces more propolis.

Also, Mizis (1978) found that each colony irrespective of race and method of collection can produce from 50-700 g propolis and that with Caucasian mountain Gray bees gathered 2-3 times more quantity of propolis.

So, it obvious that F_1 Italian bees is more suitable to collect propolis in Egypt than F_1 Carniolan bees. (Fig.5)

Table (3): The amounts of propolis gathered by two races of honeybee during a year of study (from January to December, 1986), in grams /colony/year.

Colony No.	F ₁ Italian bees	F ₁ Carniolan bees
1	92.95	45.95
2	61.15	52.65
3	63.65	44.25
4	71.40	43.55
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Total	289.15	186.4
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Mean	72.29	46.6

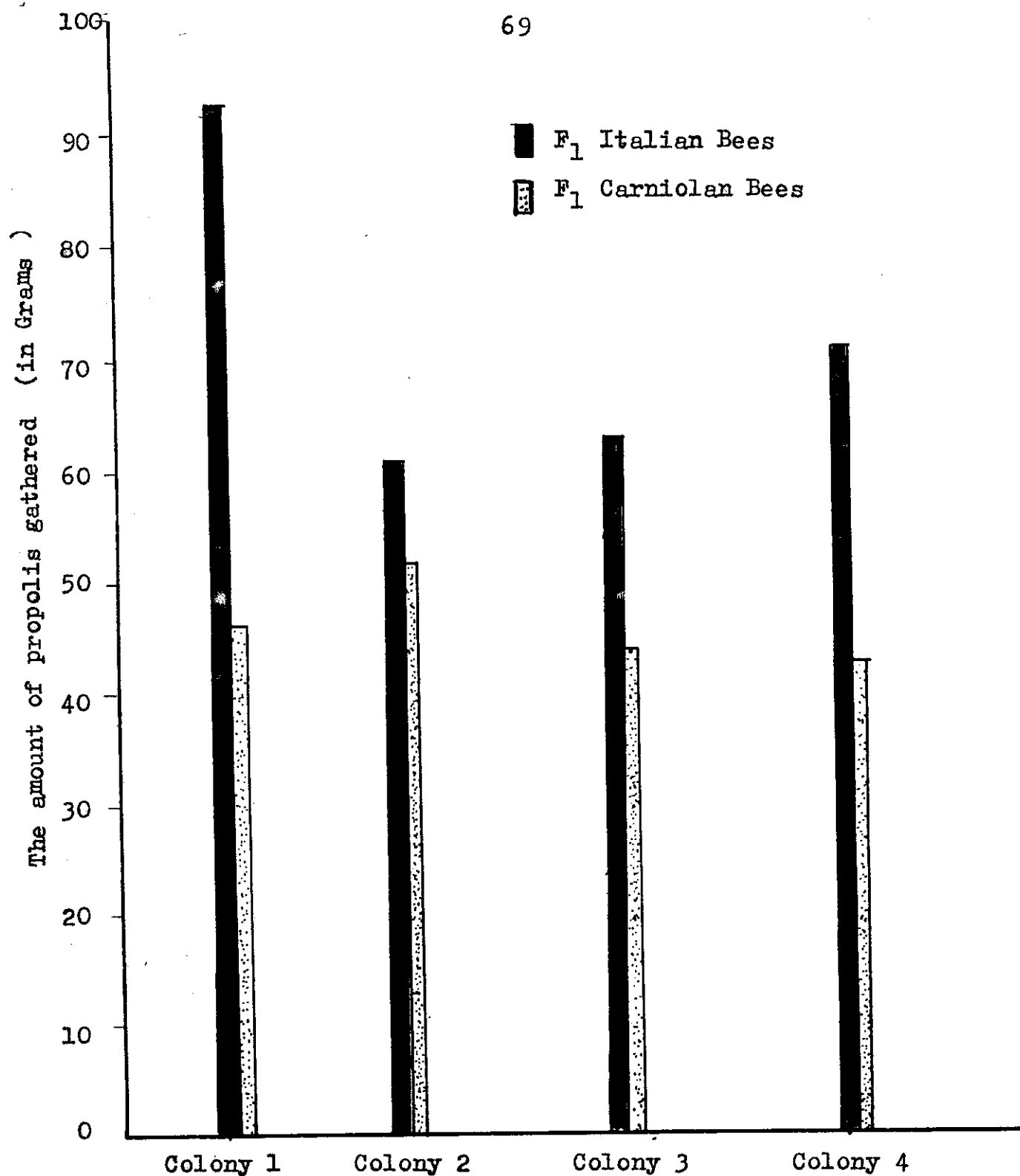


Fig. (5) : The amounts of propolis gathered by the two races of honeybees during a year (1986).

D. Propolis gathering activity during nectar seasons:

Data in Table (4) indicates that the amount of propolis collected during citrus season which started from 15 March till 20 April, 1986 were 6.48, 8.33, 5.77 and 6.88 grams/colony for F_1 Italian bees, while were 5.77, 5.55, 3.71 and 4.61 grams/colony for F_1 Carniolan bees. In clover season (1st May - 15 June) the amounts of propolis represented 11.48, 10.40, 7.90 and 8.23 grams/colony for F_1 Italian colonies, while were 6.20, 7.53, 8.10 and 8.53 grams per colony for F_1 Carniolan bees, respectively. The amounts of propolis collected from colonies during the cotton nectar flow (1st July - 1st September) were 44.40, 22.75, 22.20 and 18.90 grams/colony for F_1 Italian bees, while were 18.65, 12.00, 4.80, and 11.25 grams/colony for F_1 Carniolan bees, respectively.

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The above results shows that the amount of propolis collected during citrus season was the lowest amount, while the clover season represent the second amount of propolis gathered, the cotton season recorded the highest yield of propolis collection by the two races of honeybees, it's gave the mean amounts (6.87, 9.50 and 27.06 grams/colony for F_1 Italian bees, while were 4.91, 7.59 and 14.18 grams/colony for Carniolan bees, respectively. Table (4) and (Fig. 6).

Statistical analysis showed that the differences between seasons and races were highly significant (Table 2).

Table (4) The amount of propolis gathered by the two races during nectar seasons at the environmental conditions during 1986.

Seasons	F _I Italian bees				Mean	% year	F _I Carniolan bees				Mean	% year
	I	2	3	4			I	2	3	4		
Citrus season												
15 Marc-20Apr.	6.48	8.33	5.77	6.88	6.87	9.50	5.77	5.55	3.71	4.61	4.91	10.54
Clover season												
1st May-15 June	11.48	10.40	7.9	8.23	9.50	13.14	6.2	7.53	8.10	8.53	7.59	16.39
Cotton season												
1st July-1st Set.	44.40	22.75	22.2	18.90	27.06	37.43	18.65	12.00	4.80	11.25	14.18	30.43
Total amount of the year	92.95	61.15	63.65	71.40	72.29	100.	45.95	52.15	44.25	43.35	46.60	100.00

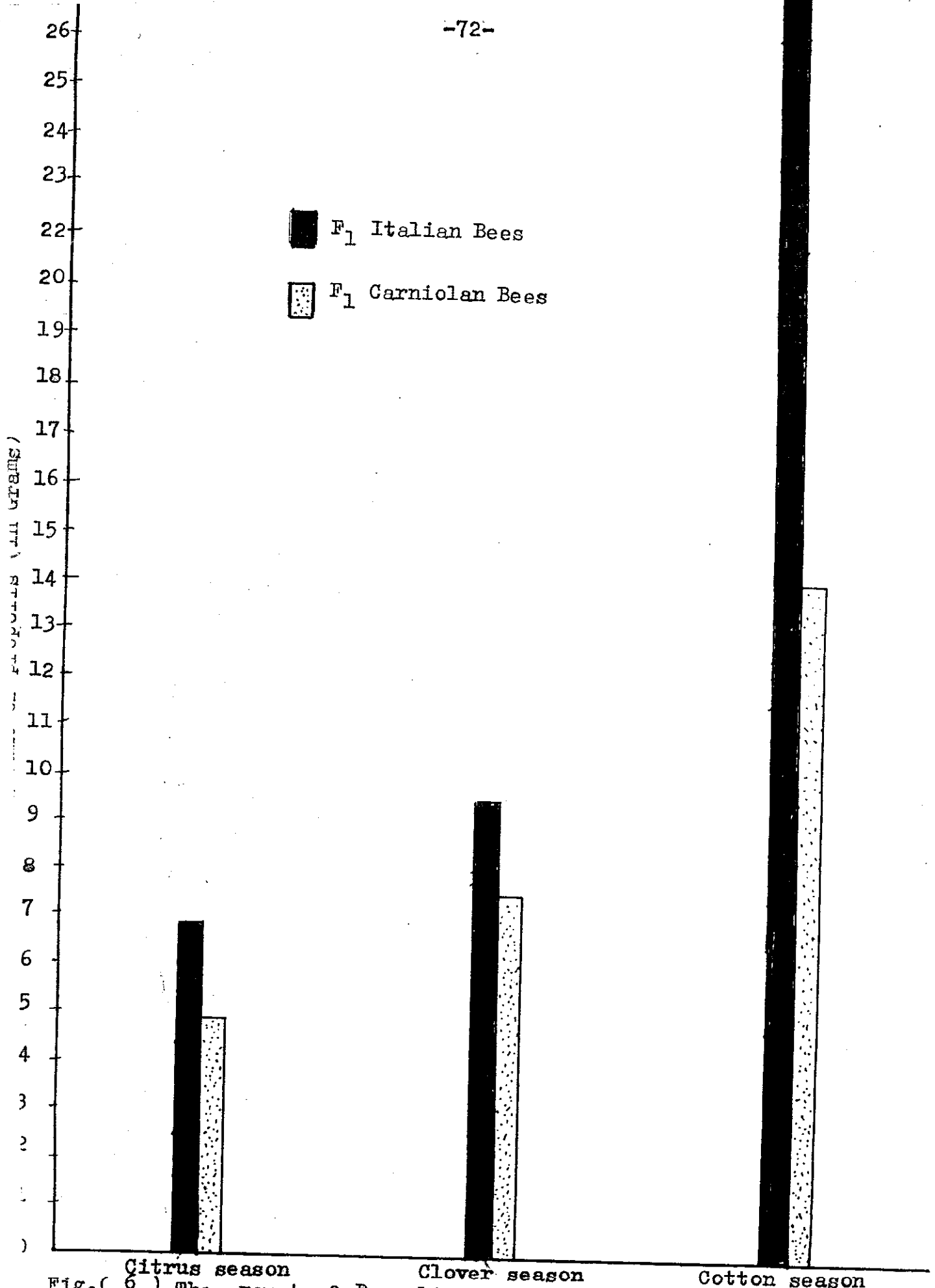


Fig. (6) The amount of Propolis gathered during the nectar-flow seasons (1986).

It can be stated from the data of the present study that the amount of propolis gathered during citrus season was less than the amounts gathered during clover and cotton seasons. That is may due to the start of the active season of honey bees, and the small population density during spring. The main aim of the colony at this time is to increase its strength. Also, during citrus season, temperature become warm and several trees, Eucalyptus, sallow, casuarina, apricot, pear and peach begin to bloom. So, honey bee workers begin to collect propolis from the sticky substances surrounding the new buds. Propolis collection during clover season is more than citrus season, for the amounts of blooming trees are more especially Eucalyptus sp. safsaf and popular and the gradual rose in atmospheric temperature which helps bee workers to collect more propolis. It is known that, cotton period extend for a long time during the summer season, high temperature, many blooming trees and vegetables help the bees to collect a big quantity of propolis. So, the results show the percentage of seasons gathering of F_1 Italian bees were 9.5%, 13.14% and 37.43% of the total amount of the year, while for F_1 Carniolan bees were 10.54%, 16.39% and 30.43% of the total amount of the year, through citrus, clover and cotton season, respectively.

It concluded that the amounts of propolis gathered by F_1 Italian bees ~~were~~ more than that collected by F_1 Carniolan bees during different seasons. These results indicated that these data are in agreement with many authors, El-Sarrag (1977),

found that Sudanese bees and their hybrids collected more propolis during the summer (9.0-12.7 g) than in winter (2.5-9.0 g), while the figures for Carniolan bees were 4.1 and 1.79 g, respectively in Egypt.

Ayoub (1982) in Egypt, who indicated that the total amount of propolis gathered in winter represented 15.2% of the total yield. In spring, the amount of propolis reached 26.3% and in summer the quantity increased to 37.0%. The same author added that in autumn propolis production was 21% of propolis production during the year.

E. The relationship between brood rearing activity and propolis gathering:

The brood-rearing activity started to estimate the amount of workers brood from 17 January till 22 December, 1986 (1 square inch = 25 worker cells).

Data represented in Tables (5 and 6) and Fig. (7) indicate that the total amount of sealed brood area of F_1 Italian colonies was 62663 inch² with an average 15640.75 inch² per colony/year. While in F_1 Carniolan colonies, the sealed brood area during a year was 48113 inch² with an average 12028.25 inch² per colony/year.

Statistical analysis of the average sealed brood area in the two races showed that F_1 Italian bees were significantly higher than F_1 Carniolan bees (Table, 7).

Data in Tables (5 & 6) show an obvious relationship between brood rearing activity and the amounts of propolis gathered by the races of honey bee. Generally speaking, sealed brood activity and the amounts of propolis gathered by F_1 Italian bees are more than F_1 Carniolan bees. There are some factors which may affect the range of propolis gathering such as temperature, humidity, and the colony needs of the hive.

Less amounts of propolis gathering and brood rearing activity were in January and December. F_1 Italian bees gathered 2.59 and 1.94 g of propolis and produced 389.50 and 209.06 in² respectively while, for F_1 Carniolan bees were

Table (5) : Sealed brood activity of workers in two strains of honeybees (P₁ Italian and P₂ Carniolian) during a year of study. (January 198 to December 1986).
(in square inches)

Date of Measurements	F ₁ Italian bees					F ₁ Carniolan bees					total
	Col.No. 1	2	3	4	Total	1	2	3	4		
January , 17	190	140	133	165	628	120	98	115	110	443	
, 29	150	165	105	130	550	111	125	180	72	488	
February, 10	380	500	550	850	2280	290	410	300	355	1355	
, 22	565	625	690	995	2875	525	480	680	610	2295	
March, 8	620	720	675	880	2895	508	514	696	540	2258	
, 20	736	815	750	1100	3401	670	568	645	759	2642	
April, 2nd	185	848	940	1054	3627	698	670	582	790	2740	
, 14	710	740	660	815	2925	480	525	605	660	2270	
, 26	795	843	830	940	3413	410	470	458	565	1903	
May, 8	690	530	500	635	2495	425	450	430	480	1785	
, 20	705	720	560	750	2735	485	512	520	600	2117	
June, 1st	770	730	610	890	3000	540	620	530	710	2500	
, 13	850	696	700	975	3221	532	710	640	834	2816	
, 25	670	710	920	1015	3315	605	700	670	765	2740	
July, 7	950	754	990	970	3664	750	310	745	780	3195	
, 19	310	1030	1155	1295	4300	330	920	1030	850	3630	
, 31	900	730	1020	910	3620	720	730	360	715	3093	
August, 12	680	630	595	805	2710	600	530	640	615	2385	
, 24	585	480	552	640	2207	536	410	315	435	1746	
September, 5	305	412	480	506	1703	350	280	270	317	1217	
, 17	328	365	411	420	2741	218	255	210	200	883	
, 29	370	480	312	395	1557	178	267	310	325	1090	
October, 11	225	235	205	200	865	155	124	139	210	628	
, 23	198	250	211	175	834	85	136	117	110	448	
November, 4	112	130	118	142	502	90	125	105	128	448	
, 16	95	75	88	110	368	50	45	96	78	269	
, 28	87	115	84	115	401	45	37	24	65	171	
December, 10	125	124	98	120	477	55	97	120	17	239	
, 22	90	140	105	76	411	98	106	68	48	320	
Total	14576	14847	15067	18073	62663	11369	11784	12217	12743	48104	

Table(6): Monthly average amounts of estimated brood
and propolis gathering activity .

Month	F _I Italian bees		F _I Carniolan bees	
	propolis g	brood in ²	propolis g **	brood in ² *
January	2.59	389.50	1.88	289.21
February	3.16	1555.63	2.36	1134.29
March	4.31	1967.75	2.75	1513.58
April	7.06	1736.04	4.93	1306.17
May	6.20	1746.25	4.89	1401.17
June	6.60	2754.50	5.40	1827.96
July	10.71	2338.00	6.77	2080.13
August	16.33	1477.17	7.40	1210.23
September	3.93	1270.24	2.90	611.79
October	4.41	490.40	3.08	330.58
November	5.05	670.22	2.45	363.21
December	1.94	209.06	1.80	200.21
Total	72.29	16604.76	46.60	12268.53

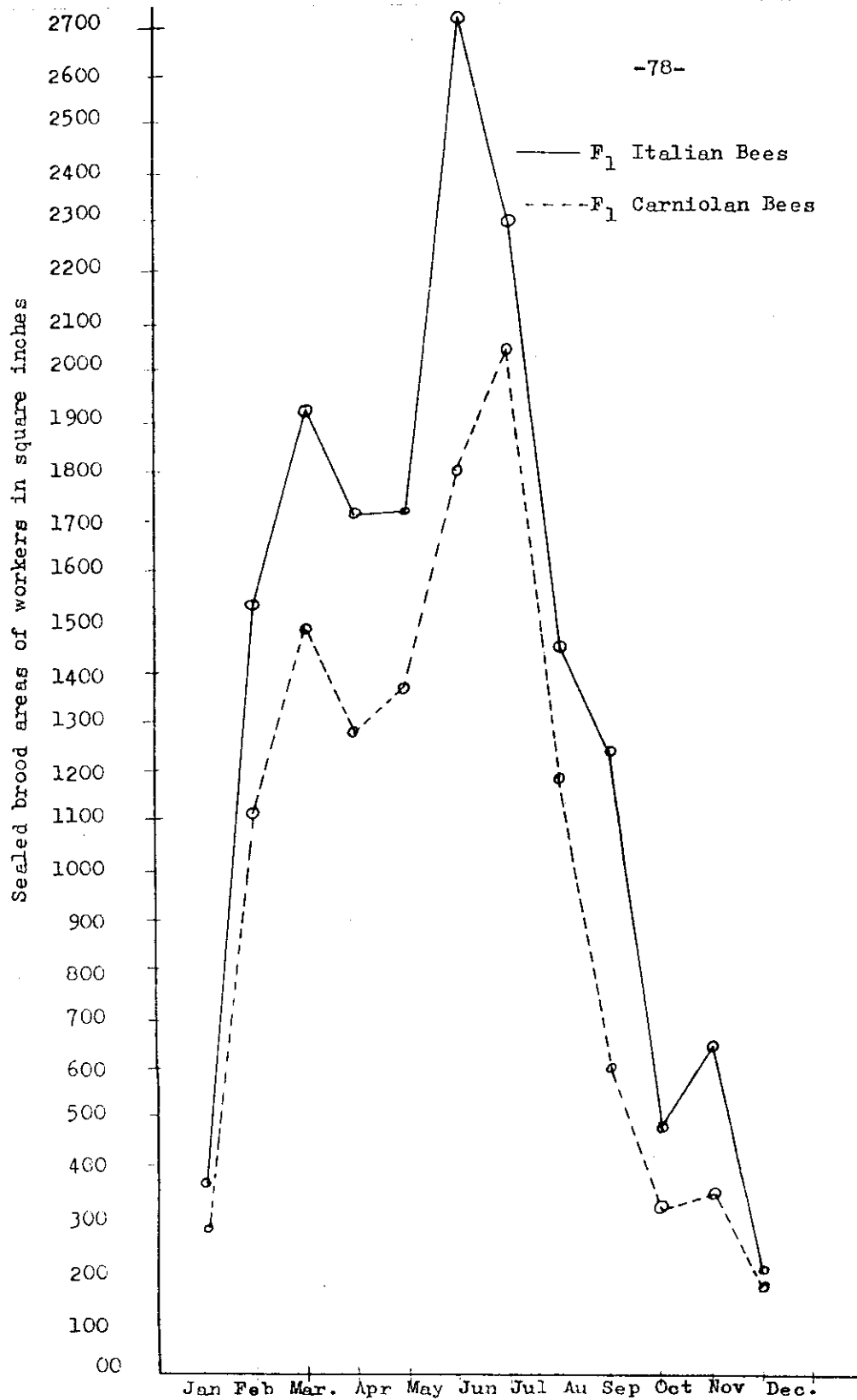


Fig. (7): The activity of honeybee on sealed-brood of workers during a year (1986)

Table (7): Analysis of variance of brood rearing of the two races during 1986.

Source variation	D.F.	S.S.	M.S.	F
Colonies	3	12120.31		
Races	1	900010.78	900010.78	7.39
Dates	28	19552501.80	698303.64	5.73

Error	199	24238093.48	121799.46	

Total	231	22126019.38		

* Significant.

** Highly significant.

1.88 and 1.80 g of propolis and 289.21 and 200.21 in² respectively. This means that through dearth months, honey bee

In Spring, the amount of gathered propolis and sealed brood areas of F₁ Italian bees were 19.09 g and 5981.62 inch², while by F₁ Carniolan bees were 14.17 and 4432.89 inch².

In summer, the amount of gathered propolis and sealed brood areas of F₁ Italian bees were 31.59 g and 5530.69 inch², while, by F₁ Carniolan bees were 18.10 g and 4337.86 inch². It can be stated that, the high level of propolis gathering and the high activity of producing brood occurred in spring and summer. This may be attributed to the facts that, in spring, honey bees start the activity with the warm temperature, and beginning of blooming flowers. At this time of the year, honey bees increase their population and at the same time the sources of propolis are available. In summer, prevailing high temperature help the bees for breaking off the pieces of the resinous exudate from the buds, by using their hind legs and mouth parts, while it is hard and difficult to gather propolis in cold weather. Therefore, the amount of propolis gathering, is more in summer than in spring. Table (8) and Fig. (8).

In autumn, honey bees gather propolis less than spring but more than winter. Also, sealed brood production of F₁ Italian bees was 1698.49 inch² and the weight of propolis 12.32 g. The same trend was observed in F₁ Carniolan bees

Table (8) : Season amounts of propolis gathered and estimated sealed brood activity
by the two races during a year of study.

Season	F ₁ Italian Bees		F ₁ Carniolian Bees		
	Sealed brood (in) ²	Propolis (g)	Propolis (%)	Sealed brood (in) ²	Propolis (g)
Spring	5981.62	19.09	26.41	4432.89	14.17
Summer	5530.69	31.59	43.70	4337.86	18.10
Autumn	1698.49	12.32	17.04	995.92	7.60
Winter	3393.86	9.69	13.40	2501.86	6.55
Total	16604.66	72.29		12268.53	46.42

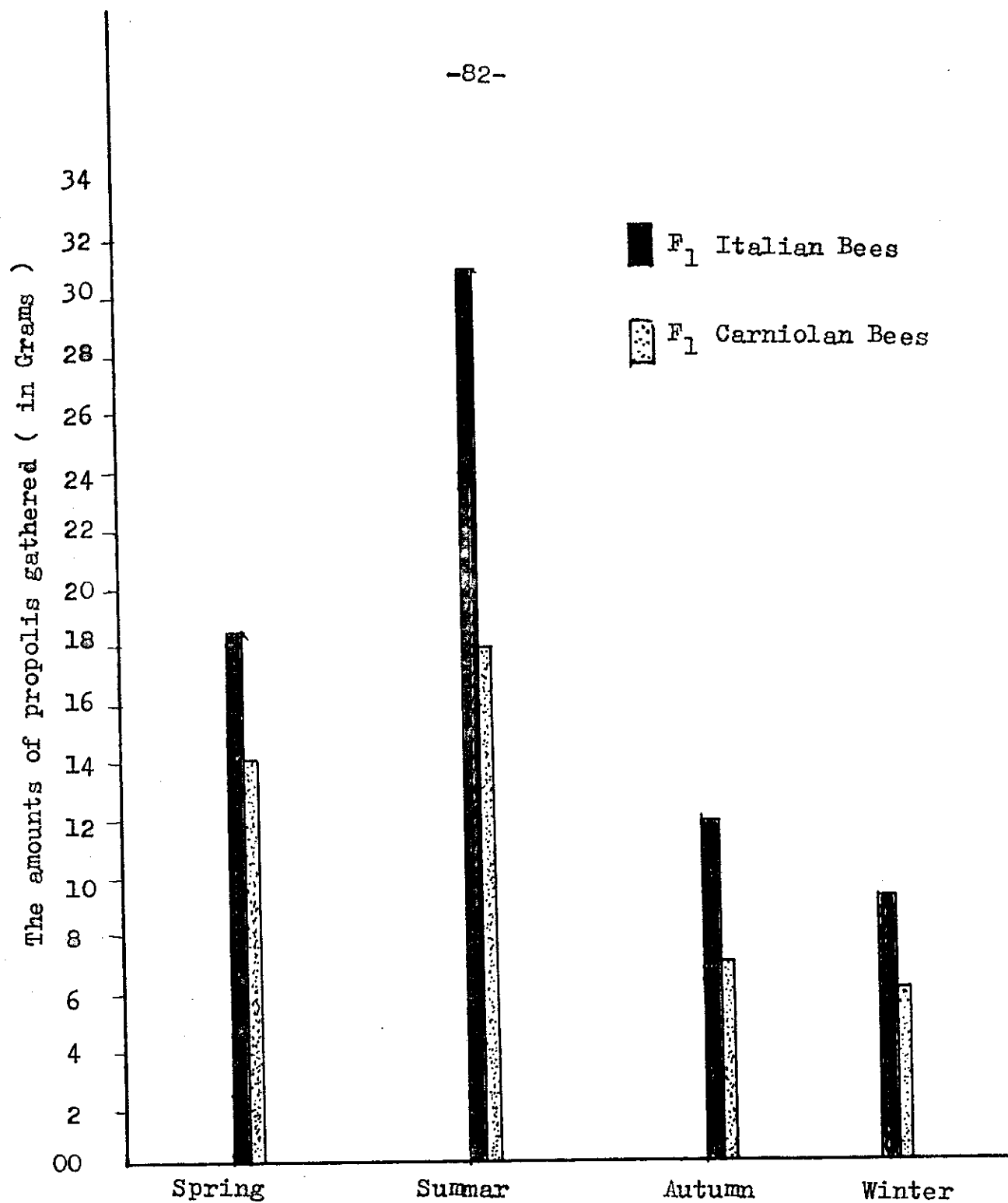


Fig.(8) : The amounts of propolis gathered during different seasons (1986) by the two-races of honeybees.

which gathered 7.6 g propolis and the area of sealed brood area was 995.92 inch².

In winter, the lowest level of gathering propolis and sealed brood rearing activity were recorded. Cold weather, Lazy honey bees and small population density and the physical properties of propolis which became hard in low temperature result in gathering small amounts of propolis. F₁ Italian been gathered 9.69 g propolis with brood rearing activity of 3393.96 inch². Meanwhile, F₁ Carniolan bees gathered the lowest amount of propolis, i.e. 6.55 g with 2501.86 inch² of brood rearing area.

From the above data, it can be concluded that F₁ Italian bees collect more propolis and rear more brood than F₁ Carniolan bees. The amounts of propolis and the areas of sealed brood can arranged in a descending order from summer to spring to autumn and then to winter.

It concluded that the amounts of propolis gathering activity during different seasons represented F₁ Italian hybrid 26.41%, 43.70%, 17.04% and 13.40% for Spring, Summer, Autumn and Winter respectively. While F₁ Carniolan bees were 30.53%, 38.99%, 16.37% and 14.11% for 4 seasons, respectively. These results were in agreement with Ayoub (1982) who indicated that the total amount of propolis gathered in winter represented 15.2% of the total yield. In spring, the amount of propolis

reached 26.3% and in summer the quantity increased to 37%. The same author added that in autumn propolis production was 21% of propolis production during the year.

El-Sarrag (1977) found that Sudanese bees and their hybrids collected more propolis during the summer (9.0-12.7 g) than in winter (2.5-9.0 g) while the figures for Carniolan bees were 4.1 and 1.79, respectively.

P. Chemical Composition of Propolis:

In this experiment we used the effect of different solvents in extraction on fractionation of the chemical constituent of Egyptian crude propolis. Thus the crude propolis have been fractionated by different solvents such as water, ethyl alcohol, light petroleum ether (80-100°C) benzene and acetic acid, then identified the chemical constituents of each fraction. The method of extraction can be summarized in the following:

1. Water Extraction:

The extracted^{anting} obtained by boiling a crude propolis with water after removed the bees wax, the filtrate was dissolved in ether and the ethereal layer partitioned three times with 10% NaOH solution. Evaporation the ethereal layer (neutral fraction) gave a compound crystallise from 50% aq-ethanol as Pale yellow waxy material (I), melted at 62-64° which identified as beeswax melting point and mixed melting point determination and also spectroscopically.

Infrared spectrum of the compound (I) showed bands at 722-728 cm^{-1} (C-C skelton), at 1175 cm^{-1} (C-O, at 1730 and 1463 (S CH), at 1740 cm^{-1} (C-O) and stretching at 2860-2925 cm^{-1} (CH aliphatic) Fig. (1). Thus infrared spectrum is in agreement with Merck index (1983).

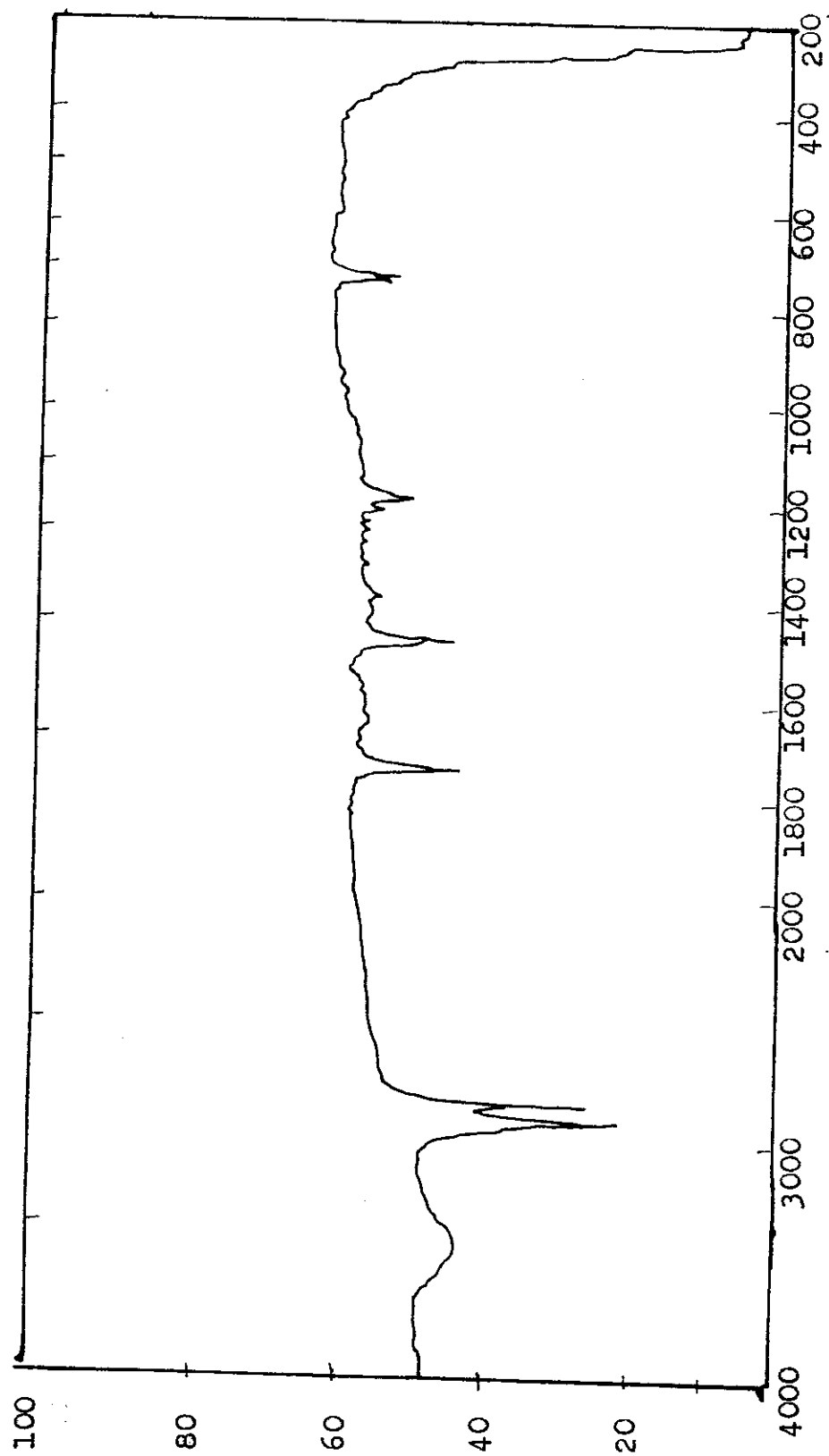


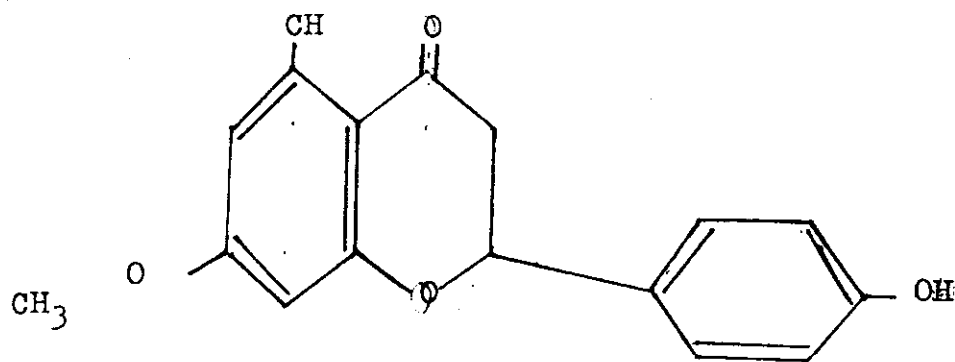
Fig. (10) : Infrared spectrum of water extraction of propolis (WEP).

The nuclear magnetic resonance (NMR) spectrum of the above compound (in dimethyl sulfoxide) showed a multiple signal at value 1.0-1.5 protons of (CH_3 and $-\text{CH}_2-$) and broad signals centered at 3.5 (for protons at $\text{CH}_2-\text{CO}-\text{CH}_2$) and absence any signal attributable to olefinic and aromatic protons. (Fig. 9)

Acidification the sodium hydroxide fraction by dil. hydrochloric acid (5%) then extracted by ether to give a very small compound (2), crystallized from aq. ethanol (70%) and melted at 92° (yield 0.02%) and a brown color compound (3) crystallized from ethanol as main product melted at $150-152^\circ$. The structure of (3) was confirmed by comparing its melting point with (Ghisalberty, 1978).

The infrared spectrum of the compound showed attributable bands corresponding to stretching of ether linkage at ($1100-1260 \text{ cm}^{-1}$), benzenoid bands at ($1615-1420 \text{ cm}^{-1}$), bending vibration of CH at ($1340, 1450, 1460 \text{ cm}^{-1}$), stretching vibration of carbonyl group at 1690 cm^{-1} and stretching vibration of hydroxyl group as broad band ($2500-3500 \text{ cm}^{-1}$) c.f. Fig.

From all the above compound was confirmed as Sakuranetin (4,5-dihydroxy-7-methoxyflavone).



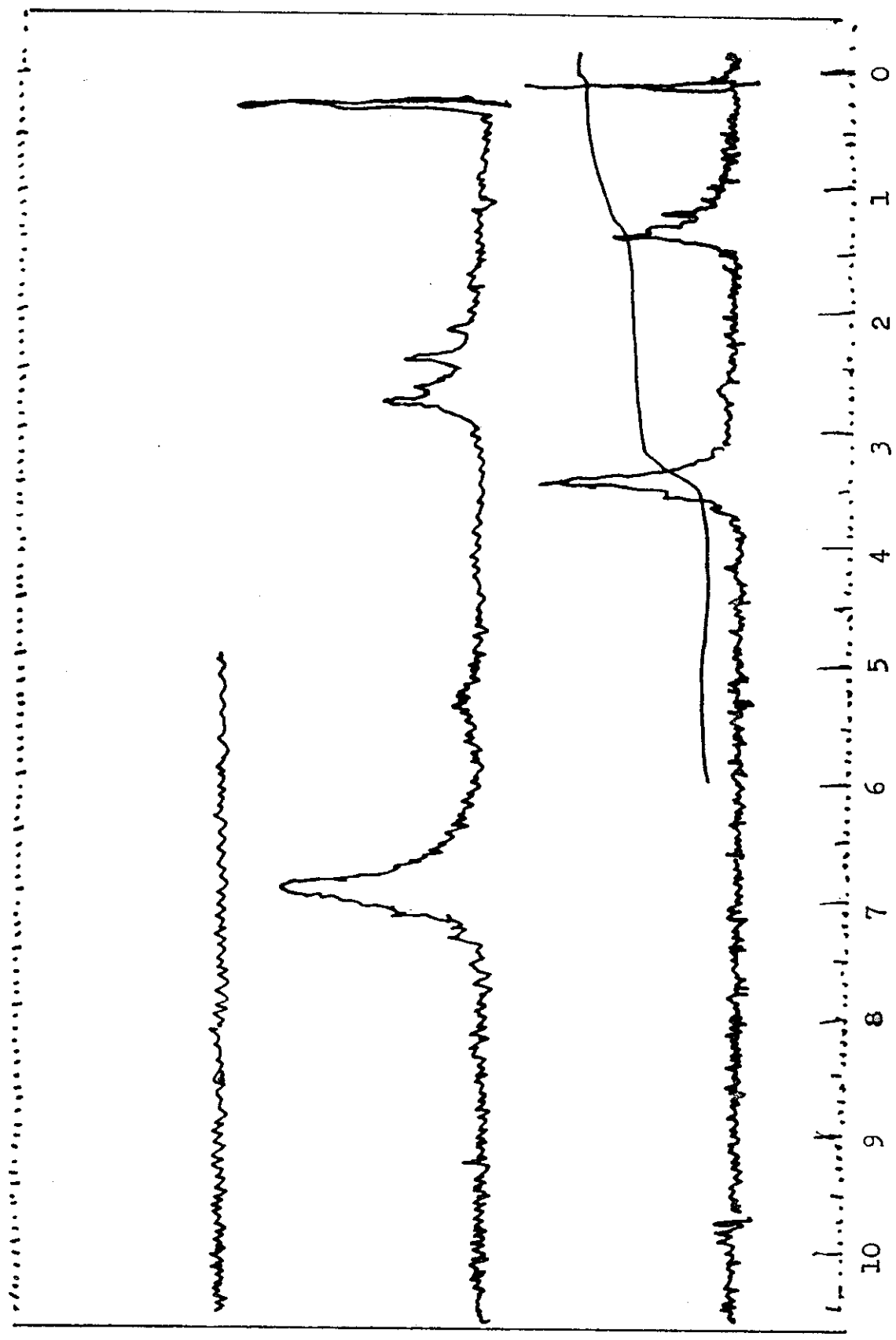


Fig. (9): N M R for Bees-Wax.

2. Propolis obtained by extraction with 70% aq. ethanol:

A. Method A.:

i. The oily matter obtained by extraction with dil HCl 5% was very small yielding and could not be identified. (0.02% by weight).

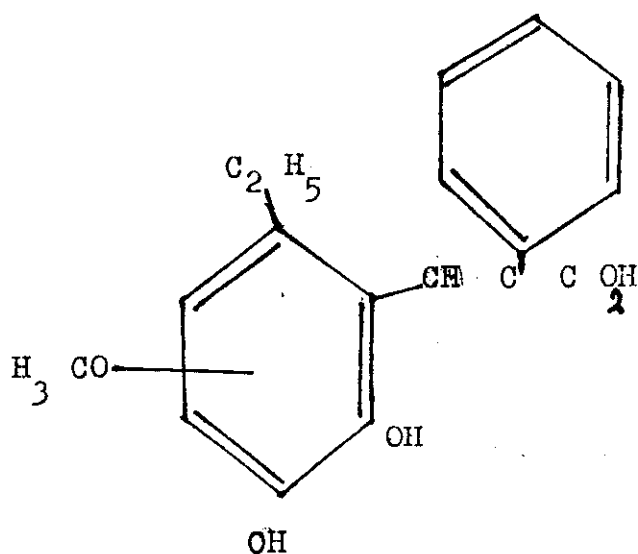
ii. The product obtained from sodium carbonate and sodium bicarbonate extraction identified as beeswax by melting point (62° - 64°) also by its infrared spectrum.

iii. The sodium hydroxide fraction after neuterlisation by HCl (5%), extracted by ether and evaporated to give a compound (4) as brown color crystallised from ethanol/benzene and melted at 158 - 162° . The structure of (4) was confirmed by:

a. The infrared spectrum of the compound (4) showed bands attributable to stretching vibration of $-O-$ (1150 cm^{-1}) bending vibration of CH of CH_3 and CH_2 groups at (1340 , 1430 cm^{-1}), stretching vibration of $\text{C} = \text{C}$ olefinic and aromatic (1600 cm^{-1}), stretching vibration of CO of COOH (shoulder at 1680 cm^{-1}), stretching vibration of CH of $-\text{CH}_2-$ and CH_3 or CH- at (2900 cm^{-1}) and stretching vibration of CH as broad band centered at (3500 cm^{-1}) c.f. Fig. (11).

b. The nuclear magnetic resonance (NMR) spectrum of compound (4) in (dimethyl sulphoxide- D_6) showed signal at 0.8 and 1.2 (broaded for 5H of aliphatic, 2.8 (s, 3H of $\text{CH}_3\text{O Ar}$) and 6.6-7.9 (m7H of aromatic and olifinic.) Fig.(12). The infrared of spectrum compound attributed hydroxy methoxy alkyl cinamic acid derivative.

90



HYDROXY METHOXY ALKYL CINAMIC ACID
DERIVATIVE

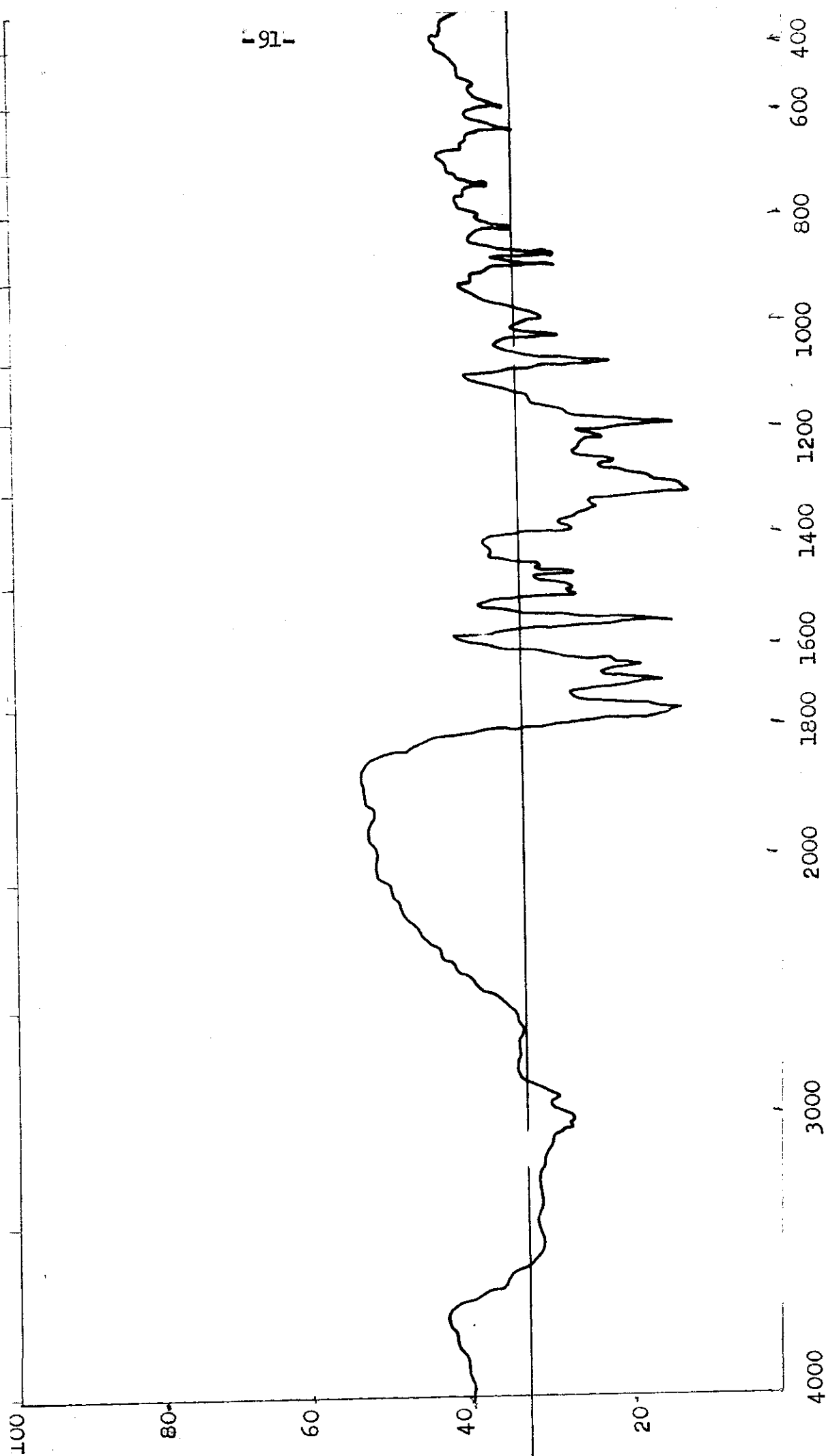


Fig (11) : Infrared spectrum of the alcohol extracted propolis compound (AEP).

Method B:

The neuteral ethereal layer of light petroleum (40°-60°), (60°-80°), (80-110° , benzene and ethyl alcohol evaporated till diyness to give oily matter 5, 6, 8 and solid compound 7 and 9 respectively. The aq-sodium hydroxide solution of the above solvents neutralic by dil. HCl, then extracted by ether and evaporated till diyness to give oily matter 10, 11, 12 and solid compound 12 respectively. The results are listed in the following table (9).

Table (9): Different compounds obtained from propolis by some solvents.

Solvent	Neutral fraction state	Alkaline fraction state
Light petroleum 40-60	5 (oil)	10 (oil)
Light petroleum 60-80	6 (oil)	11 (oil)
Light petroleum 80-110	7	12
Benzene	8 oil	13 oil
Ethyl alcohol	9	-

Compound 9 has a mp. (152-185°) and has a biological activity.

3. Soxhelt extraction:

For determined the different fractions of extracted propolis, which fractionated had a highly biological activity. This work deals with the extraction of 60 gm of crude propolis by Soxhelt using the following solvents in the same sequence,

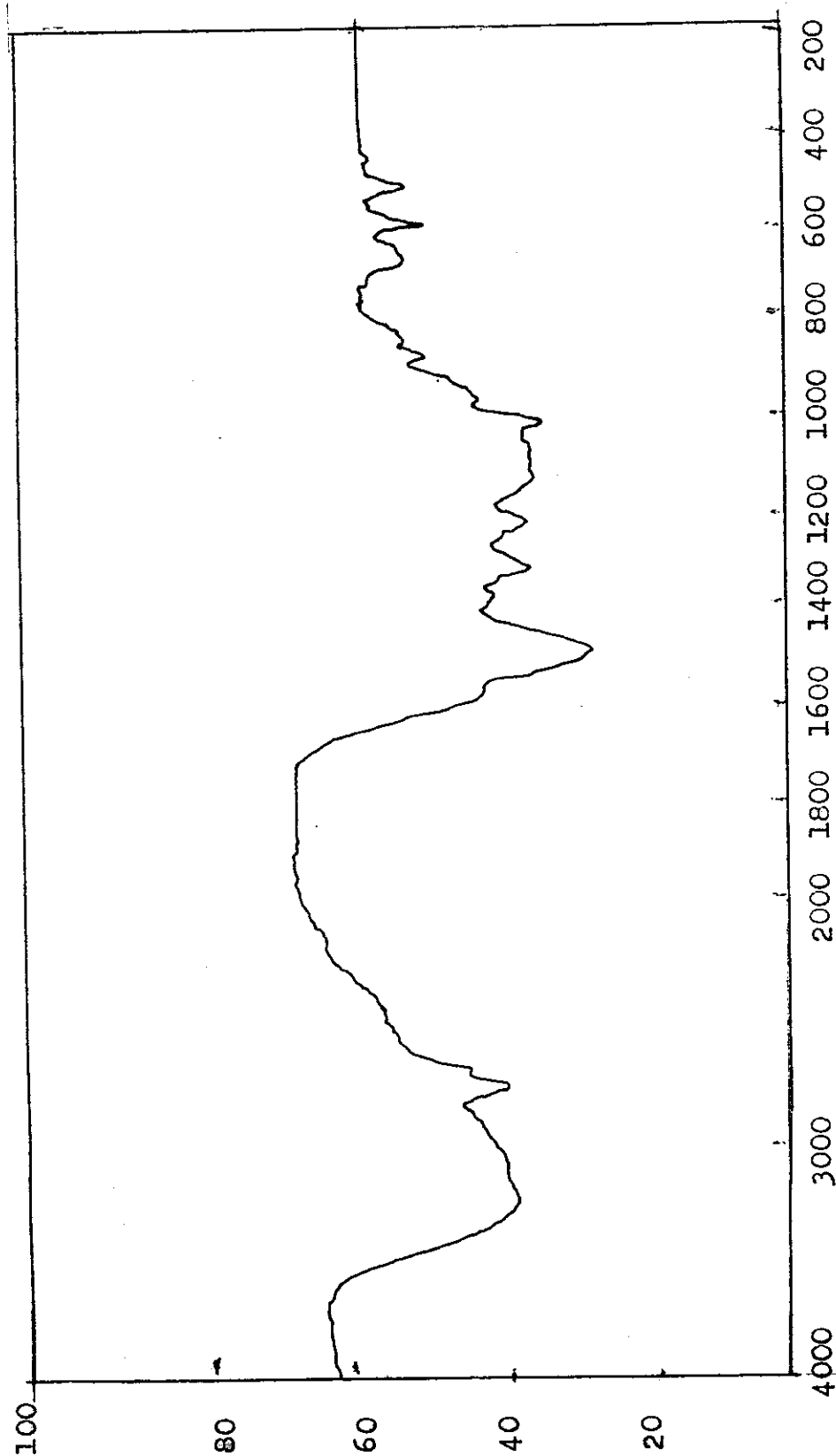


Fig. (13) : Infrared spectrum of the compounds extracted from the propolis
extracted with 70 % aq-ethanol.

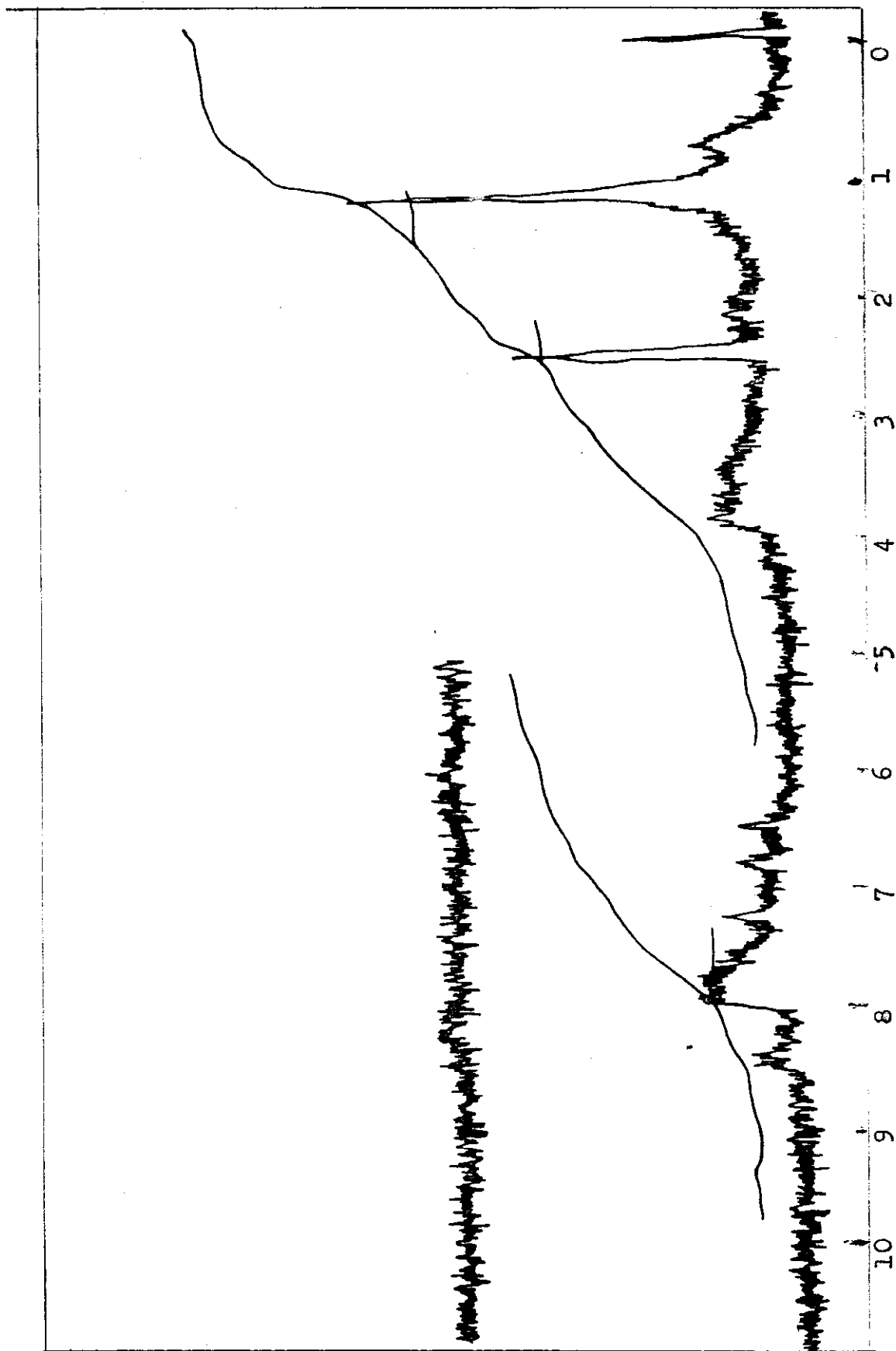


Fig. (12) : The number magnatic resonance (N M R) for propolis.

light petrol (b.p. 80-110°C), benzene, ethyl alcohol and acetic acid.

The results are given in the following table (10):

Solvent of extraction	Time of extraction	Weight %	Chemical significant
Light petrol	72 hrs.	30 g	bees wax with some volatile oil.
Benzene	100 hrs.	3.5 g	beeswax, 9-12
Ethanol	100 hrs.	11.5 g	13-14
Acetic acid	120 hrs.	4.0 g	inorganic matter
Residue	--	10 g	ash

Evaporation the light petrol extract gave a yellowish comparised melted at 62-64°C, identified by comparing the infra-red spectrum and N.M.R. as bees wax. (Fig. 13).

On further investigation, the evaporation of benzene extracted the residue obtained dissolved in ether and partitioned with NaOH evaporation the ethereal layer gave about 0.2 g yellowish compound, identified as bees wax, while acidified the aqueous layer by 5% HCl and extracted by ether. Evaporation the ethereal layer gave a brownish residue which on spotting in thin layer chromatography (T.L.C.) and eluted by light petrol (60-80°C)/benzene (V/V) appeared four spots. (fig. 14).

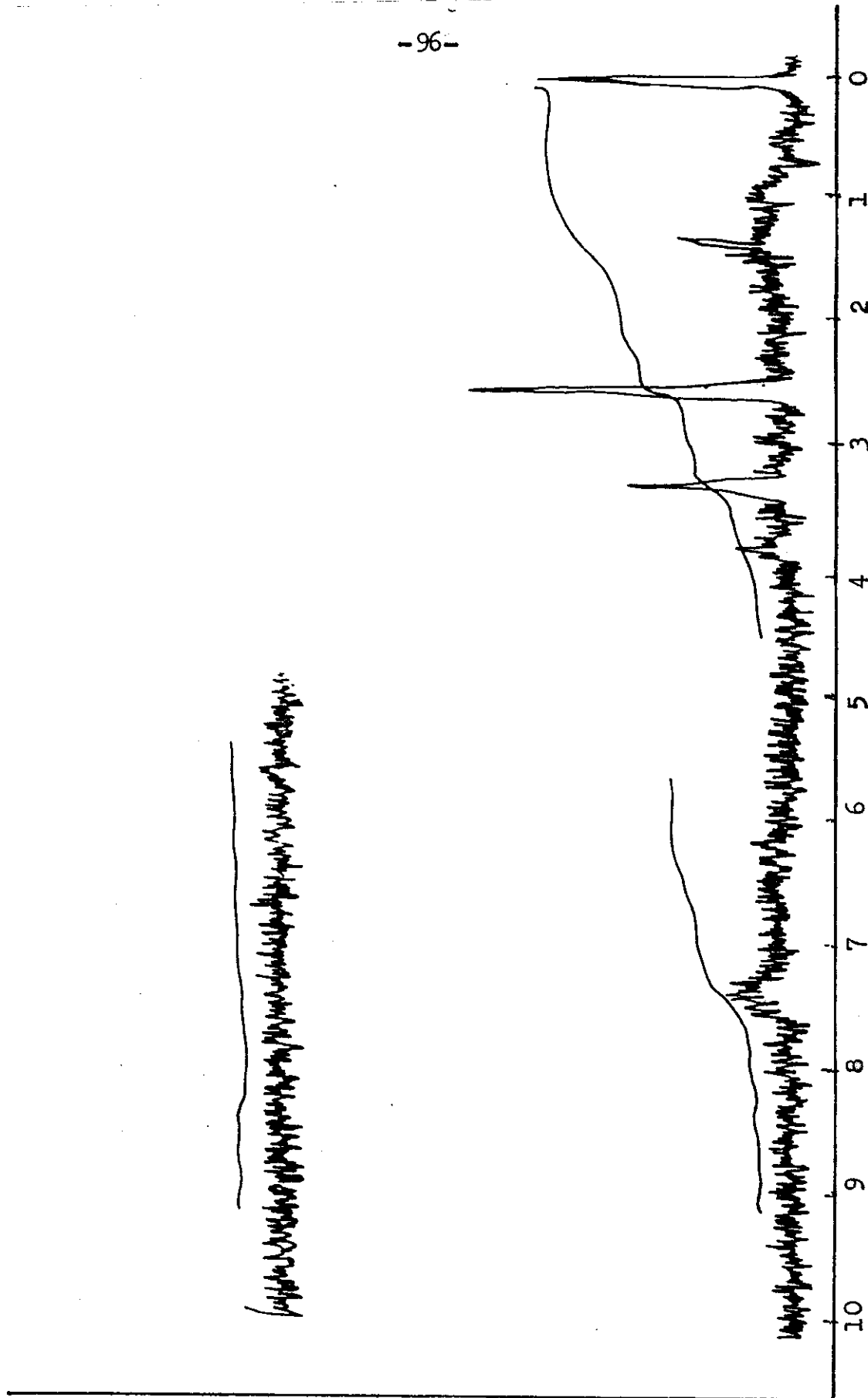


Fig. (16) : The number Magnetic Resonance (N M R) for propolis.

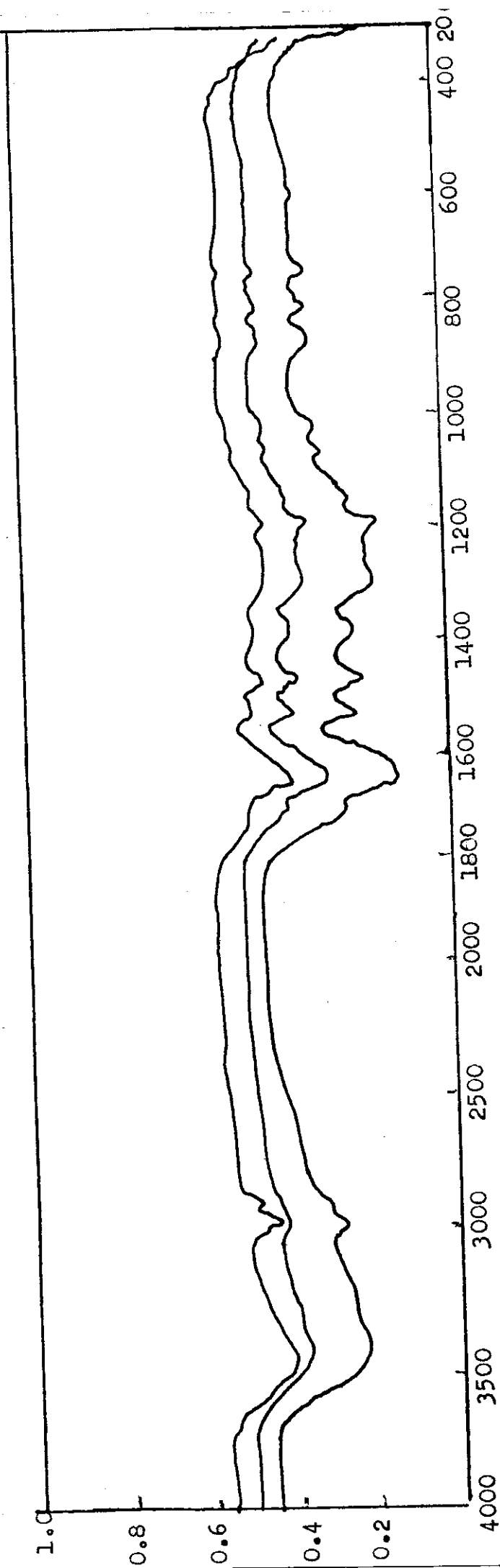


Fig. (15) Infrared spectrum ' (I R) for propolis compound.

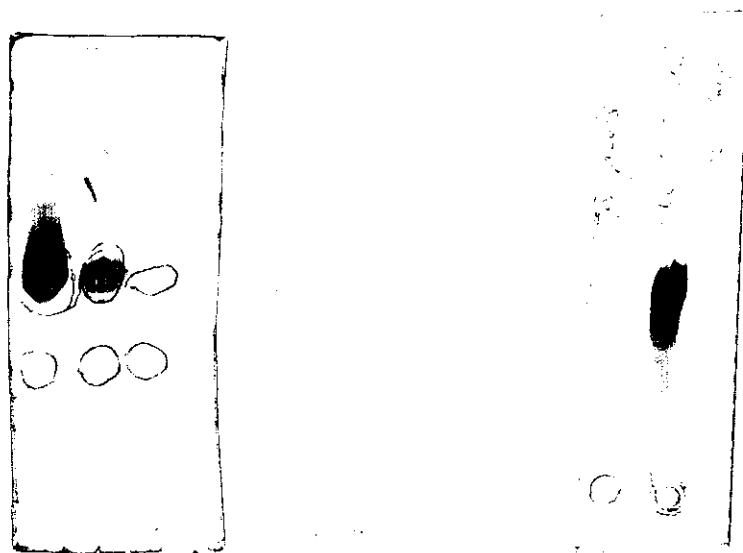


Fig. (14): Evaporation the Ethereal layer gave a brownish residue which on spotting in thin layer chromatography(T.L.C.).

The brownish residue separated on preparative T.L.C. and eluted by light petrol (60-80°°/benzene (V/V). Scratching the zones and extracted by acetone and slow evaporation gave the following compound 9, 10, 11 and 12 melted at 138 (decomposed), 148° (decomposed), 239° and over 300° respectively. The infrared spectra of 9, 10, 11 and 12 were confirmed as hydroxy-methoxy flavone derivatives Figs. (15 , 16 , 17 and 18).

Also, evaporation the alcohol extract and spotting the residue on T.L.C. showed two spots by elution with benzene/light petrol (80-110°) (3 V/V). Fractional distillation of the residue by benzene/light petrol (80-110°) gave brown color solid 13 melting point 107° and brown color solid from alcohol 14 melted at 227-230°).

The infrared spectra of 13 and 14 and N.M.R. of 13 and 14 in (DMSO-D 6) confirmed that the components 13 and 14 are 3-alkyl derivatives of hydroxy-methoxy flavones.

The acetic acid extract after evaporation gave an inorganic compounds.

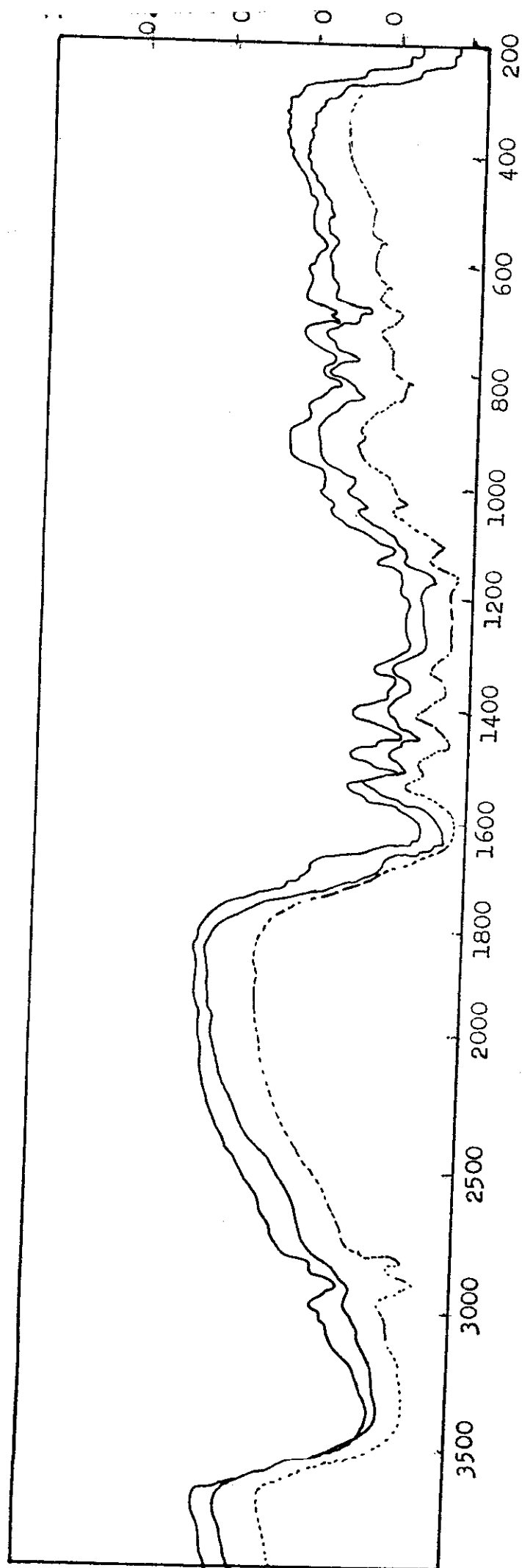


Fig. (17) : Infrared spectrum for propolis.

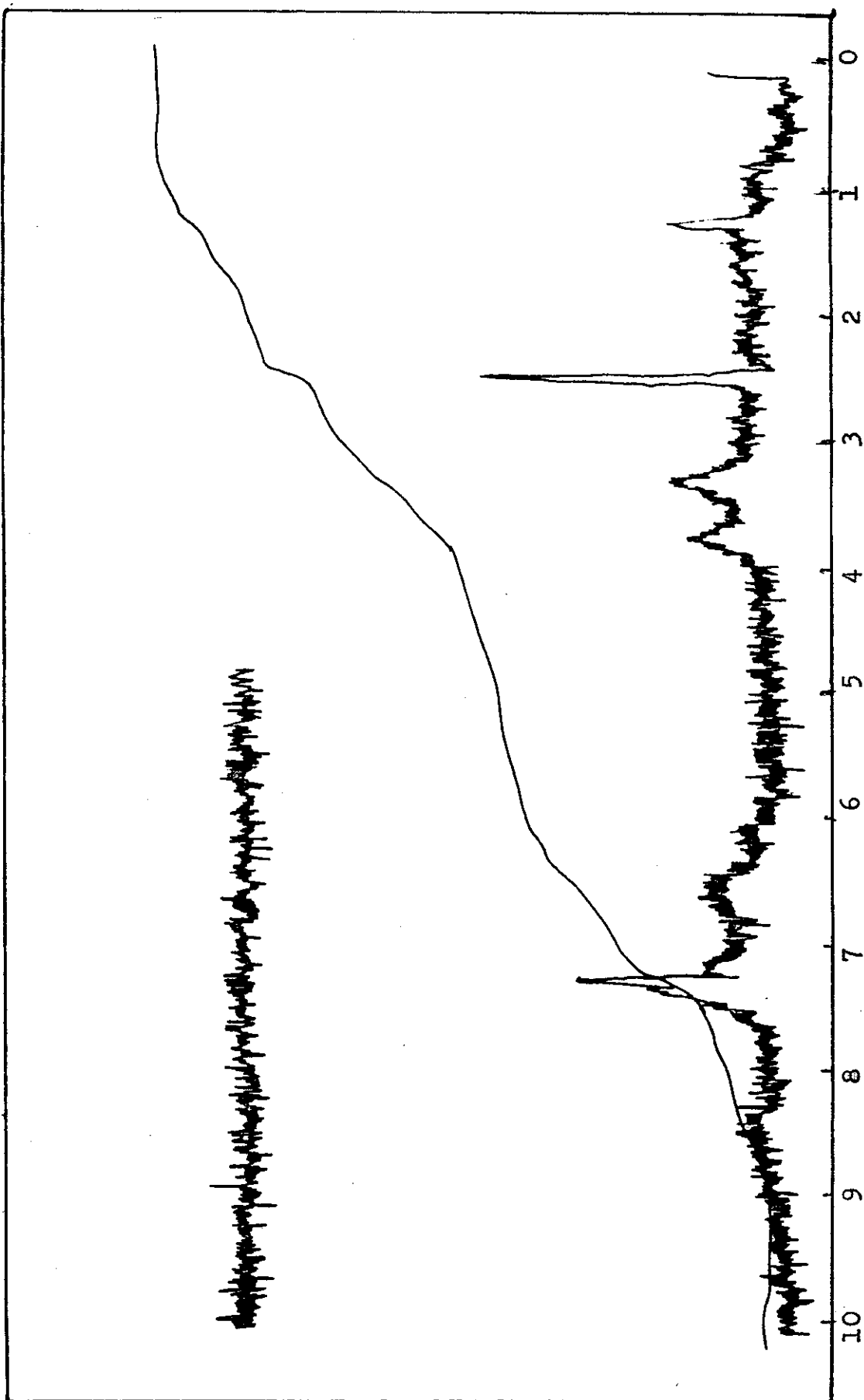


Fig. (18) : The Number Magnetic Resonance (N M R) for Propolis.

✓ G. Biological effects of propolis:

Propolis is a resin used by the bee to close hive cracks and to ensure its proof against damp and water. The second use is also very important, when the bees kill an enemy in the hive and its corps is too big to be removed. it is embalmed in propolis which reserves it from decay or subsequent mouldiness. In fact, even at that time, propolis was considered as cicatrizing and anticeptic substance, both for animal and plant tissue.

a. Antibacterial activity:

The aim of this work is to prove in vitro the anti-microbial activity of alcohol, acetone and water extract solutions from samples of Egyptian propolis which were collected by honeybees for carrying out experiments on some food poisoning microorganisms, one bacterial strain, Staphylococcus aureus as gram-positive microorganisms and five bacterial strains as gram-negative microorganisms; Salmonella typh-marium, Salmonella newport, Escherichia coli O₇₈, Escherichia coli O₁₁₁, and Proteus vulgaris.

1. Staphylococcus aureus:

By the application of the preparations, water, ethyl alcohol, and acetone extract, as bacteriostatic on vitro by using the above indicator microorganism, clearly showed that WPE has obvious bacteriostatic activity. While EPE and APE have slight bacteriostatic activity (Table 11). These results

agreed with Kivalkina (1969) who found that extracts of propolis have been shown to potentiate the effect of certain antibiotics towards Staphylococcus aureus by addition it to the nutrient medium. As well as, Chernyak (1973) found that propolis at 1.25-5 mg/ml, showed bactericidal effect towards 20 St. Vachet (1978) found that propolis has an antibiotic effect on the Gram-positive St. aureus. Also, Meresta and Merest (1983 & 1985), Cheorghieva (1987) and Maciejewicz (1987) sured the same above results.

2. Salmonella typh-marium and Salmonella newport:

The results showed that, WPE has a bacteriostatic on vitro by using the above indicator microorganisms. But APE has less marked effect towards the same microorganisms. While EPE has negligible effect as bacteriostatic. These results agreed with Lavie (1960) who found that propolis showed activity towards Proteus vulgaris and less activity towards Salmonella gallinarum, Sal. pullorum and Sal. dublin. Also, Villanveva et al. (1964) used some fractions of propolis, galangin and pinocembrin, 1.6 mg/m to inhibit Sal. gallinarum for 24 h. Again, Lavie (1978) recorded that, for propolis antibiotic, very interesting reaction on Proteus vulgaris activity reducing to half or even less on Salmonella pullorum and Sal. gallinarum.

3. Escherichia coli₇₈ and Escherichia coli₁₁₁:

It is obvious that WPE has a bacteriostatic activity on both E. coli strains, but EPE and APE have a medium activity.

Table (11): Propolis extracts effect on some food poisoning microorganisms.

Microorganism	WPE	EPE	APE
Salmonella typhimarium	+	+	-
Salmonella newport	+	+	-
Escherichia coli ₇₈	+	+	+
Escherichia coli _{III}	+	+	+
Proteus vulgaris	++	+	+
Staphylococcus aureus	++	+	+

++Obvious effect

+ Effect

- None had effect.

The effect of all extracts were less marked towards Salmonella. These results like exactly with Lavie (1960) who found that propolis negligible activity towards different strains of E. coli, but Lavie (1978) showed zero reaction on four strains of E. coli. Ghisalberti (1979), Cheorghieva (1987) recorded that the extracts of propolis have antibacterial effect in vitro.

4. Proteus vulgaris:

Water propolis extract showed bacteriostatic activity towards Proteus vulgaris. Alcohol-propolis extract and acetone-propolis extract were less marked towards Proteus vulgaris.

As well as Lavie (1960 & 1978) found that propolis showed bacteriostatis activity towards Proteus vulgaris.

b. Antifungal activity:

The obtained different propolis fractions, Sakurantín, Cinamic acid derivative and flavon were subjected to study their biological effect on fungi. The fungus Alternaria was used as test organism in this work.

Data presented in table (12) show clearly that the biological effect of the different tested propolis fractions was greatly conspicuous. The fraction Sakurantín clearly fungicidal effect. This fraction Sakurantín did not affect percentage of spore germination in the low concentrations, but it clearly decreased the length of germ-tube with increasing its concentrations. It completely inhibited both spore germination and germ-tube growth at concentration of 960 ppm. Further studies of this fraction about its fungal toxicity must be carried out.

Concerning the biological activity of both fractions cinamic acid and flavone, the data in table (12) indicated that both fractions showing growth promotion properties. The germ-tube growth of Alternaria spores were considerably increased with increasing concentration of these fractions, until the concentration of 480 ppm then begin to gradually decreased. Further studies are needed to detect the promotion effects on these compounds on different organisms.

These above results are in agreement with Cizmarik and Trupl (1975) who found that ethanolic extract of propolis has

Table (12): Biological assays of different propolis compounds fractionated from different solvents on percentages of spore germination and germ tube length in u of Alternaria sp. after 16 hours from incubation at 25°C.

Concentrations in ppm	Sakurantin			Cinamic acid			Flavone		
	Germ %	Germ- tube length u	Germ %	Germ tube length u	Germ %	Germ tube length u	Germ %	Germ- tube length u	Germ- tube length u
Control	96	160.9	96	160.9	96	160.9	96	160.9	160.9
30	96	125.6	94	161.6	94	161.6	96	185.2	185.2
60	95	103.5	95	180.4	95	180.4	97	201.7	201.7
120	95	84.5	96	207.2	96	207.2	99	240.5	240.5
240	96	74.5	97	216.8	97	216.8	99	262.0	262.0
480	86	68.9	99	249.4	99	249.4	99	290.0	290.0
960	00	0.0	99	259.6	99	259.6	99	282.0	282.0
1920	00	0.0	99	256.2	99	256.2	96	280.0	280.0

a fungicidal activity. Also, they found that the alcoholic extracts inhibited about 38 strains of skin fungi. Herman ((1983) recorded that an extract of propolis was found to be effective against a number of strains of fungi. Pepelijnjak et al. (1985) found that the pure propolis extracts, a concentration of 15-30 mg/ml was needed to inhibit the growth of Candida albicans, Aspergillus flavus, A. achraceus, Penicillium vividicatum and P. notatum.

C. Effect of Propolis Extracts on *Broala coeca* attached on honeybee queens:

Table (13) indicates the numbers of honeybee lice *Broala coeca* on honeybee queens. All propolis extracts showed quickly effect, where lice on queens disappeared after half an hour of the treatment of spraying. The examination of honeybee queens treated with propolis extracts showed the number of lice on queens were not more than 25% of initial numbers on these queens. It means that using 10% of water propolis extract, acetone propolis extract and ethanol propolis extract (as spraying on honeybees castes will be reduced the number of lice until the seventh day from application. On comparison these results obtained from treating with different propolis extracts with control (distilled water) showed that WPE reduced the number of lice on queens as represented 11.11%, 8.33% and 0.00% of initial numbers. In case of EPE represented 9.09%, 20.00%, and 10.0% of initial numbers and APE represented 25%, 25% and 11.11% of initial numbers, respectively, While using distilled water application showed that no changes on the infestation with bee-lice on the queens these results represented 111.11%, 83.33% and 100% of initial numbers.

It is obvious from the above results that the application of any propolis extracts especially WPE gave a good result on reduced the bee-lice infestation which parasite on queens. Many authors used some chemical control methods for

Table (13) : Effect of different propolis extracts on Braula coeca in honeybee colonies

Propolis extracts	Repli- cates.	Initial numbers on queens	Hour after treatment				Days after treatment				Final Number for last day	
			1	2	1	2	3	4	5	6	7	(%)
Water Ext.	1	9	-	-	-	1	2	1	2	1	1	11.11
	2	12	-	-	-	1	1	1	1	-	1	8.33
	3	8	-	-	1	1	-	1	1	1	-	0.00
Ethyl Alcohol Ext.	1	11	-	-	1	1	2	2	2	1	1	9.09
	2	10	-	-	1	1	1	1	2	2	2	20.00
	3	10	-	-	1	1	1	1	1	-	1	10.00
Acetone Ext.	1	8	-	-	-	1	1	1	1	1	2	25.00
	2	12	-	-	1	1	2	2	2	2	3	25.00
	3	9	-	-	1	1	1	-	1	1	1	11.11
Distilled water												
(Control)	1	9	9	9	9	10	9	11	9	9	10	111.11
	2	12	11	11	10	10	11	11	10	10	10	83.33
	3	9	9	9	9	9	8	7	9	8	9	100.00

treating bee-lice (tidion, folbex and phenothiazine). These chemicals found to be caused some resual effect which that effect on honeybee castes but in our methods of lice control will be enhanced this trend of application the colonies for pests control.

These results are in agreement with Ayoub (1982) who found that the treated the diet of bee-wax larvae with 10%, and 50% of propolis retardation in growth of larvae.

On the other hand Dukov (1964); Morse (1980) and Bailey (1981) who mentioned that bee-lice are usually considered harmless inquilines, but they may become pests in some areas. Daily collections from a single queen have totalled 371, about 30 being found at any one time. They used tedion, folbex and phenothiazine for chemical control.

We carried out to using the crude propolis as fumigation method by smoking the honeybee colonies with some the inner cloth's which coating with propolis. Results indicated that some honeybee workers after smoking for 5 minutes under closing the colonies were like an~~a~~esthetized without effect on bee-lice.

D. Effect of propolis extracts and its main fraction

"flavonoid" on germination of some seeds:

In this experiment water, ethanol and acetone extracts of crude propolis were used to estimate their effect on broad bean and wheat seed germination. Also, the main fraction of propolis known as flavonoid was also used for studying its biological activity on seed germination.

Table (14) indicates the effect of propolis extracts on seed germination.

a. Effect of propolis extracts on germination percentage of wheat:

It can be stated from table (14) that:

1. Ethanol propolis extract and acetone propolis extracts had an inhibiting effect on seed germination by using the different concentration throughout the germination period.

2. Water propolis extract had inhibitory action on the first stages of germination, then, germination percentage increased throughout its late period. (Fig. 19).

These results may be due to decomposition of the water propolis extract by the effect of some exerted enzymes from the seeds, but the reverse occurred by using ethanol and acetone extracts.

3. Flavonoid fraction shows inhibitory effect at concentration of 4000 and 2000 ppm in descending order, but by using 1000 ppm germination percentage was higher than in the control.

Germination percentages of control after 4, 6, 8 and 10 days were 47%, 71%, 81% and 25% while for 1000 ppm of flavonoid fraction these values were 47%, 86%, 96% and 96%, respectively. So, we notice that the low concentration of flavonoid fraction increased the germination rate.

Effect of propolis on seedling's growth of wheat:

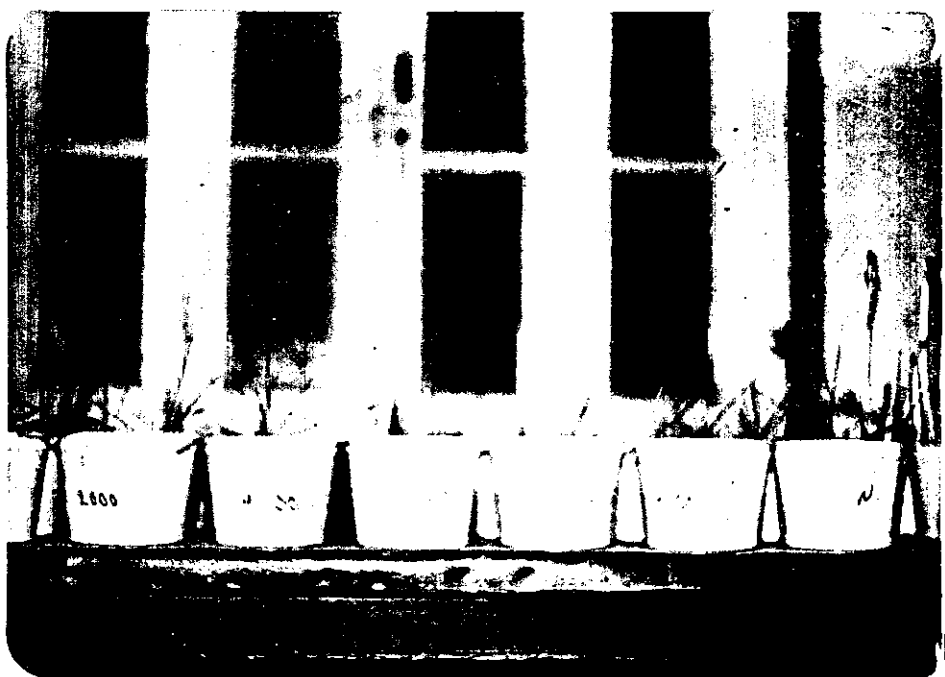
Table (15) shows the effect of different concentrations of flavonoid fraction on seedling length. This length was 21, 18.4 and 15.15 cm by using concentrations of 1000 ppm, 2000 ppm and 4000 ppm of flavonoid fraction, respectively; while for control it was 16 cm. The length of seedlings were 17.9, 11.7 and 9 cm when WPE, EPE and APE extractions were used. The above results emphasize the stimulating effect of flavonoid fraction at 1000, 2000 and 4000 ppm respectively and WPE.

b. Effect of propolis on germination of broad bean seeds:

Table (16) shows the inhibitory effect of different propolis extracts especially acetone propolis extract. On 4th day of germination, control represent 87% bean seeds' germination reached 87%, while by using water, ethanol & acetone propolis and the flavonoid fraction at concentrations of 1000, 2000 and 4000 ppm germination percentage of broad bean reached 70%, 33%, 30%, 53%, 70% and 70% respectively. The variations between percentage of germination in control and the different extracts decreased during the late period of

germination. Table 17 shows that water propolis extract stimulates seedling growth, as seedling length in this case reached 46.9 cm while the seedling length of the control was only 42.7 cm.

(Fig. 19)



(Fig. 20) : Effect of propolis extracts and flavoniods fraction on germination.

Table (14): The effect of propolis on germination percentage of wheat seeds.

Dates post germination.	Control	% seed germination by using:				
		WPE	EPE	APE	Flavonoid extract 1000	2000
4th day	47	18	28	--	47	42
6th day	71	61	39	10	86	67
8th day	81	85	47	52	96	71
10th day	85	90	53	57	96	71
						67

WPE : Water propolis extract.

EPE : Ethanol propolis extract.

APE : Acetone propolis extract.

Table (15): Mean length of wheat seedlings in cm at the 10th day of experimentation.

Treatment	Control	WPE	EPE	APE	1000Flavonoid extract	2000	4000
Seedling's length/cm	16	17.9	11.7	9	21	18.4	15.5

Table (16): The effect of propolis on germination percentage of broad bean seeds, (Vicia faba):

Dates	% of seed germination by using:				
	Control	WPE	EPE	APE	Flavonoid extract
					1000 2000 4000
<u>4th</u>	87	70	33	30	53 70 70
<u>6th</u>	100	73	47	43	80 94 83
<u>8th</u>	100	97	83	60	87 100 83
<u>10th</u>	100	97	100	70	97 100 100

Table (17): Mean length of broad bean seedlings.

Treatment	Control	WPE	EPE	APE	Flavonoid extract
Seedling's length/cm					1000 2000 4000
	42.7	46.9	27.6	23.9	35.5 31.9 34.3

II. Beeswax:

1. The amounts of beeswax and propolis found in different ages of combs:

Three honeybee combs of one year, two years, three years and over three years old were used in this study to estimate the amounts of propolis, beeswax and ecdysis skin in different ages of combs.

Table (18) shows that the mean weight of combs of one year old was 115 g. Extraction gave 91.16 g/comb beeswax, while the amount of propolis was 1.84 g/comb, which represents about 79.27 and 1.6% of its weight respectively. The moulting skin weighed 21.14 g/comb, which is about 18.38%.

Also, combs which were used for two years old, three years old and over three years gave an average weight of 119.99, 346.20 and 455.00 g respectively. Also, extractions for these different age combs resulted 91.35, 166.93 g, and 180.79 g beeswax/comb, respectively, which represents about 77.13, 48.22% and 39.43%, respectively of the comb's weight. The amounts of propolis in these different age combs were 4.14, 16.27 and 29.20 g/comb, representing 3.45%, 4.7% and 6.42% of comb's weight. The moulting skin weighed 24.54, 161.21 and 234.63 g/comb, these combs representing 20.45%, 46.57% and 51.07% of its weight. (Fig. 21).

From the above data, it is clear that the amounts of propolis increase in old combs than new combs. This relation

Table (13): The mean amounts (in grams) bee wax produced from combs of different ages.

Age of combs	Mean weight of comb	Bees wax W.	% Bees wax	Ecdysis W .	% Ecdysis	Propolis W.	% Propolis
One year	115.00	91.16	79.27	21.14	18.38	1.84	1.60
Two years	119.99	91.35	76.13	24.54	20.45	3.14	2.62
Three years	346.20	166.93	48.22	161.21	46.57	16.27	4.70
More than three years	455.00	180.79	39.43	234.63	51.07	29.20	6.42

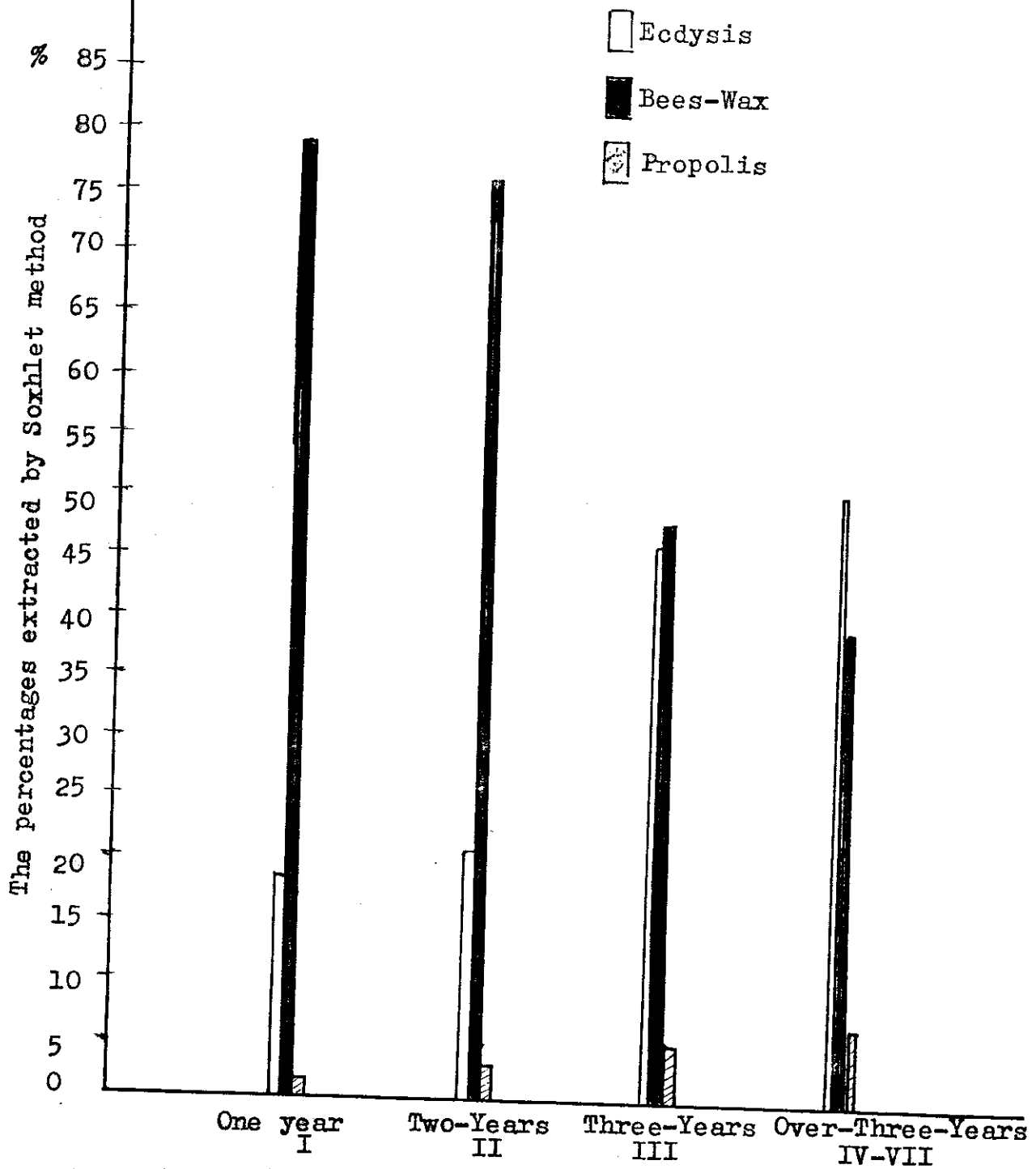


Fig. (21): The percentage of bees-wax , propolis and ecdysis from different ages of combs.

obvious that polishing by propolis for cells repeating many times with the use of combs by honeybees? Weight of moulting skin as well as increased continuously by use of the combs. The percentages of propolis and moulting skin increase in the four different ages of combs, propolis 1.6%, 2.62%, 4.7% and 6.42% respectively, while moulting skin were 18.38, 20.45%, 46.57% and 51.07%, respectively.

On the other hand, the percentage of beeswax decrease from one year old to two, three, and over three years old as it represented 79.27%, 76.13%, 48.22% and 39.4% respectively of the comb's weight. Although weight of beeswax increase generally as present 91.16, 91.35, 166.93 and 180.79 g respectively.

2. Estimating propolis and bees wax amounts in different levels of the old combs:

Data in Table(18) show that the upper level of the combs contain higher amounts of bees wax and less amounts of propolis and ecdysis. A constant weight (20 g) for every sample, three samples from each comb in three levels, upper, middle and lowest level. As mentioned before, weight of propolis in the upper level of combs one, two, three and over three years old were 0.29, 0.56, 0.66, and 1.12 g respectively, represented 1.45%, 2.8%, 3.3% and 5.60%, respectively. While the amounts of bees wax in the same samples were 16.2, 15.15, 13.44 and 12.19 g, represented 81%, 75.75%, 67.75%, and 60.95%, respectively.

On the other hand, the middle level in the same combs, contain higher amounts of propolis and ecdysis with less amounts of bees wax. (Fig. 22).

Data showed the amounts of propolis were 0.49, 0.80, 1.16 and 1.37 g represented 2.45%, 4.0, 5.8% and 6.85%, respectively. On the same trend ecdysis weights were 3.88, 4.40, 11.27 and 12.15 g, respectively.

But bees wax decreased continuously as following in the same samples 15.38, 14.66, 7.36, 6.20 g represented 76.9, 73.3, 36.6 and 31.0%, respectively.

The lowest part of combs contained less amounts of bee wax than the upper part but more than the middle part. Generally, this part of comb contain amounts of propolis and ecdysis more than the upper part but less than middle part.

As mentioned before, in Tables (18 and 19) high amounts of bees wax could be obtained again from the old combs. Mean weight of new bee foundation is about 80 g. Mean weight of new bee wax in comb three years old is 166.93 g and in comb over three years old is 180.79 g.

Simply, we solve the problem of reducing bee wax in our country. Where we obtain two bee foundation sheets from one comb three years old and more from one comb over three years old.

I hope, to have more studies for designing an instrument by using a cheap solvent or reconsidering the solvent for using it again.

Table (19): Mean amounts of bees wax and propolis in different parts of the combs (20 g. sample).

Comb's age	Part	Area cm ²	Bees wax g.	Propolis g.	Ecdysis g.	Bees wax %	Propolis %
One year	Upper	84	16.20	0.29	3.35	81.00	1.45
	Middle	75	15.38	0.49	3.88	76.90	2.45
	Lower	80	15.98	0.21	3.80	79.90	1.00
Two years	Upper	72	15.15	0.56	4.13	75.75	2.80
	Middle	64	14.66	0.80	4.40	73.30	4.00
	Lower	71.25	14.87	0.71	4.42	74.35	3.55
Three years	Upper	44	13.49	0.66	5.82	67.75	3.30
	Middle	26	7.36	1.16	11.27	36.80	5.80
	Lower	30	8.08	1.00	10.85	40.40	5.00
Over three years	Upper	30	12.19	1.12	6.58	60.95	5.60
	Middle	22	6.20	1.37	12.15	31.00	6.85
	Lower	28	6.61	1.36	11.17	33.05	6.80

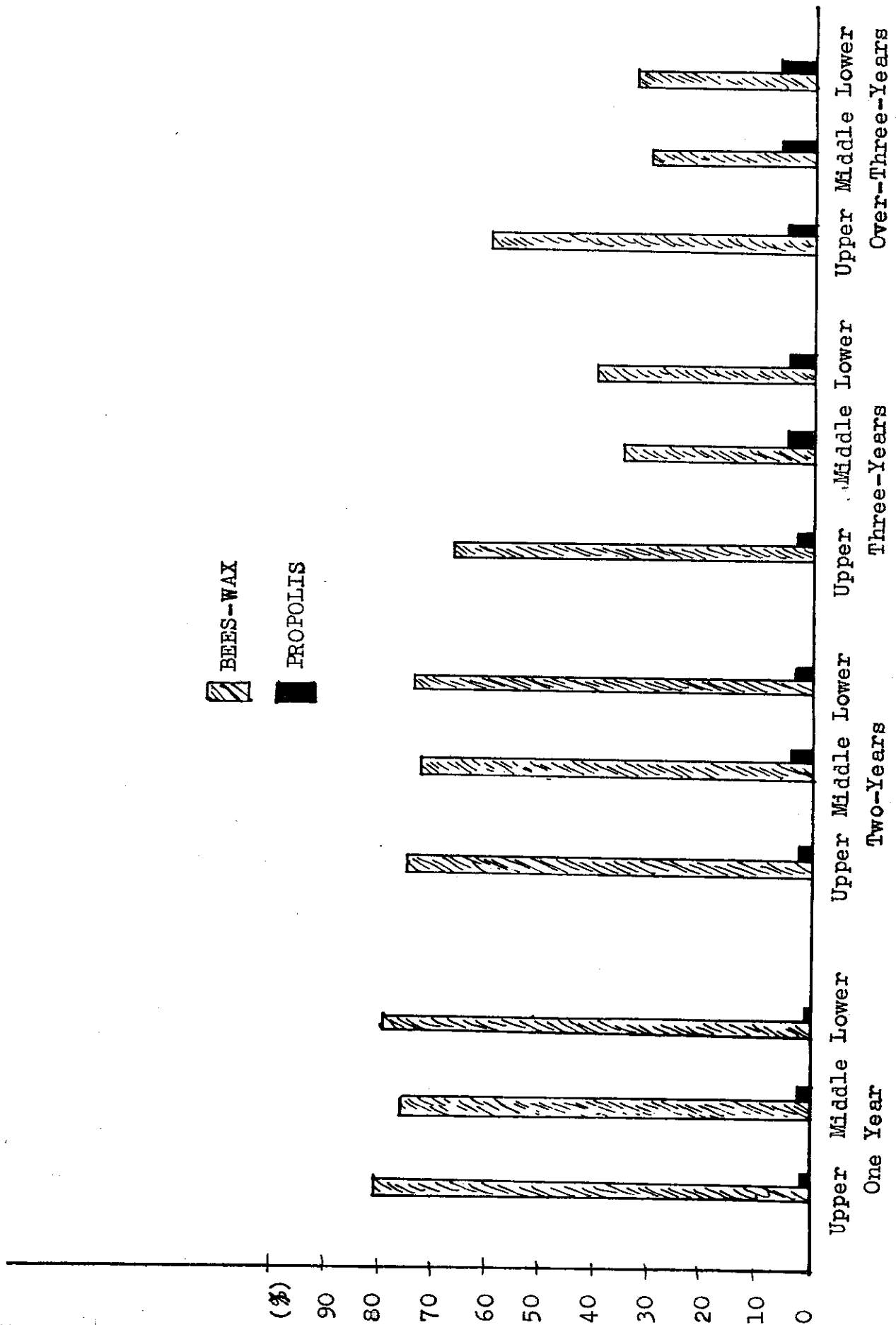


Fig. (22) Effect of different cites on the amounts of bees-wax and propolis in combs.

III. Pollen:

1. Pollen gathering activity:

Data in Table (20) Fig. (23) show that pollen gathering activity by honey bees during a year monthly. It is obvious that the largest amount of pollen gathered during June and July when clover and corn pollen were available. F_1 Italian bees gathered 105.83 in² pollen in January. On January, there is little sources of pollen, broad bean and some vegetables. On February, 584.04 in² of pollen was gathered to spread the flowers of deciduous trees (apricot, peach and pear) by F_1 Italian, 373.25 in² of pollen by F_1 Carniolan during the same month. Citrus pollen on March and April (major pollen) were gathered, 932.38 and 996.75 in², by F_1 Italian bees and were 709.46 and 751.29 in², respectively. On May, clover pollen is the main source. So, the amounts gathered by the two races were 943.06 and 618.81 in², respectively. The highest amount of pollen gathering was on June when clover pollen and corn pollen are available especially corn. F_1 Italian collected 1030.82 in² and F_1 Carniolan collected 858.32 on June. As well as, on July and August corn pollen were collected by the two races 980.38 and 926.86 in² by F_1 Italian and 706.08 and 522.98 in², respectively. Rest months, September, October, November and December, the amounts of pollen decreasing, according to the available pollen from vegetable, ornamental and weed plant by the two races.

Table (21): Analysis of variance of pollen stored in combs
of F₁ Italian and F₁ Carniolan bees during 1986
year.

S.V.	Source of variation	D.F.	S.S.	M.S.	F
Races	1	280989.1	280989.1	111.91	111
Dates	28	4164147.8	148719.6	59.23	111
Replicate	3	308670.5			
Error	199	499678.0			
Total	231	5253485.4			
111 Highly significant					

Table(20):Monthly amounts estimated of stored pollen and propolis gathered by the two races .

Months	F _I Italian bees		F _I Carniolan bees	
	polln	propolis	pollen	propolis
January	105.83	2.58	74.5	1.88
February	584.04	3.16	373.25	2.36
March	932.38	4.31	709.46	2.75
April	996.75	7.06	751.29	4.93
May	943.06	6.20	618.81	4.89
June	1030.82	6.60	858.32	5.40
July	980.38	10.71	706.08	6.77
Augst	926.86	16.33	522.98	7.40
September	411.75	3.93	248.48	2.90
October	236.40	4.41	175.19	3.08
November	195.50	5.05	86.29	2.45
December	98.31	1.94	43.17	1.80
Total	7442.08	72.29	5167.82	46.60

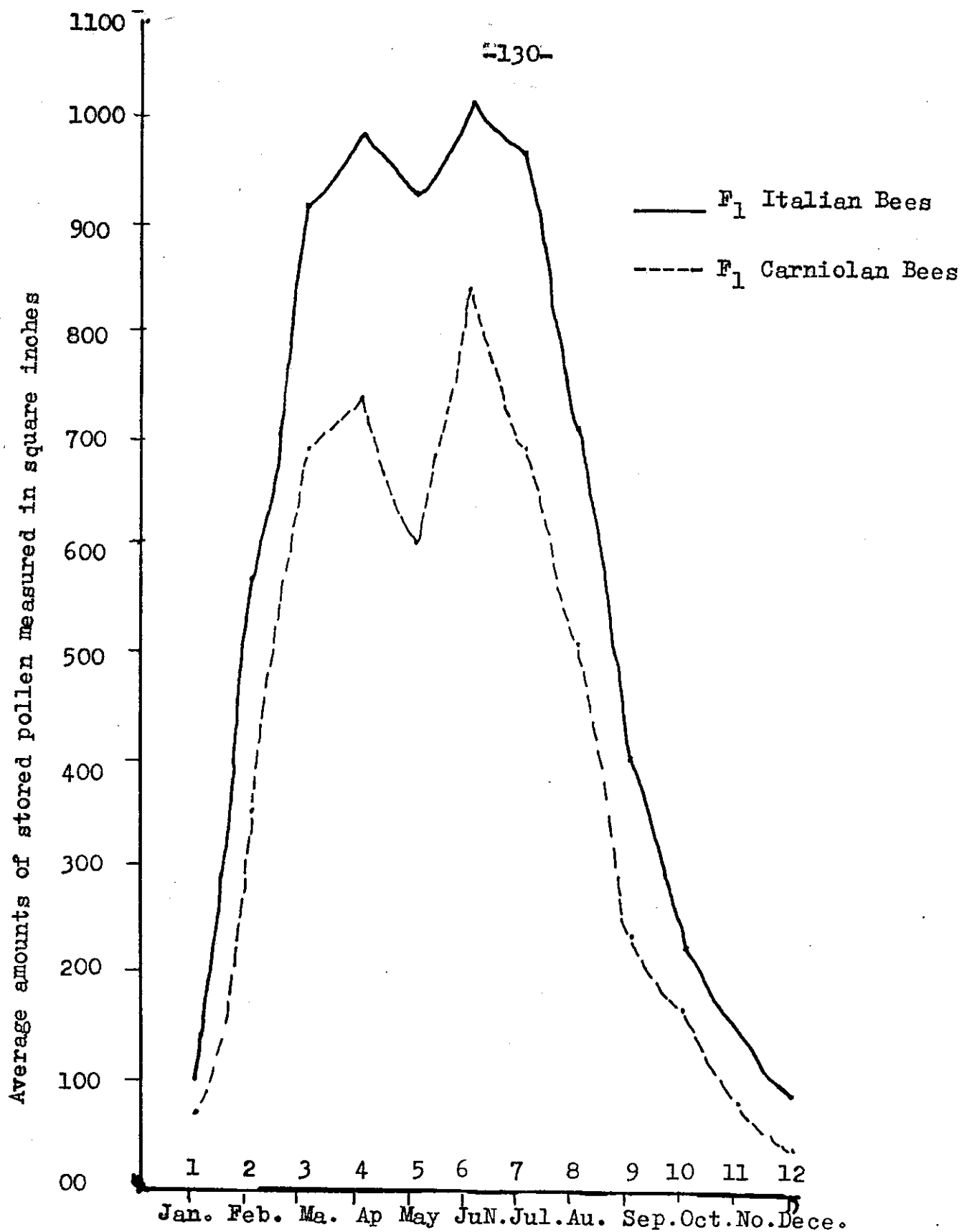


Fig. (23) : The activity of honeybees-races on stored pollen durin a year (1986).(Average in In²).

3. The relationship between pollen gathering activity and propolis collection activity:

Data represented in Table (22) showed the amounts of pollen in square inches and propolis in grams every season during a year of study.

In Spring, P_1 Italian bees collected the largest amount of stored pollen 2946.02 inch², and 18.69 g of propolis, while F_1 Carniolian bees collected 2191.21 in² of stored pollen and 14.17 g of propolis.

In Summer, F_1 Italian bees stored 2473.75 in² of pollen, and 31.19 g of propolis, while F_1 Carniolan bees stored 1630.37 in² of pollen and 18.10 g of propolis. In these two seasons the largest amounts of both pollen and propolis were collected. It is due to increase in temperature, strength of colonies, and the sources of pollen grains and propolis from different plants as above mentioned with Table (22).

In Autumn, the dearth period, less amounts of pollen and propolis were collected during a year. F_1 Italian bees and F_1 Carniolan bees gathered 572.57 and 356.11 in² of pollen and 11.92 and 7.60 g of propolis respectively. (Fig. 24).

Table (22): Amounts of pollen in square inches and weight of propolis in grains gathered by the two races during the different seasons of the year 1986.

Season	F ₁ Italian		F ₁ Carniolan	
	Pollen	Propolis	Pollen	Propolis
Spring	2946.02	18.69	2191.21	14.17
Summer	2473.75	31.19	1630.37	18.10
Autumn	572.57	11.92	356.11	7.60
Winter	1413.74	9.29	990.63	6.55

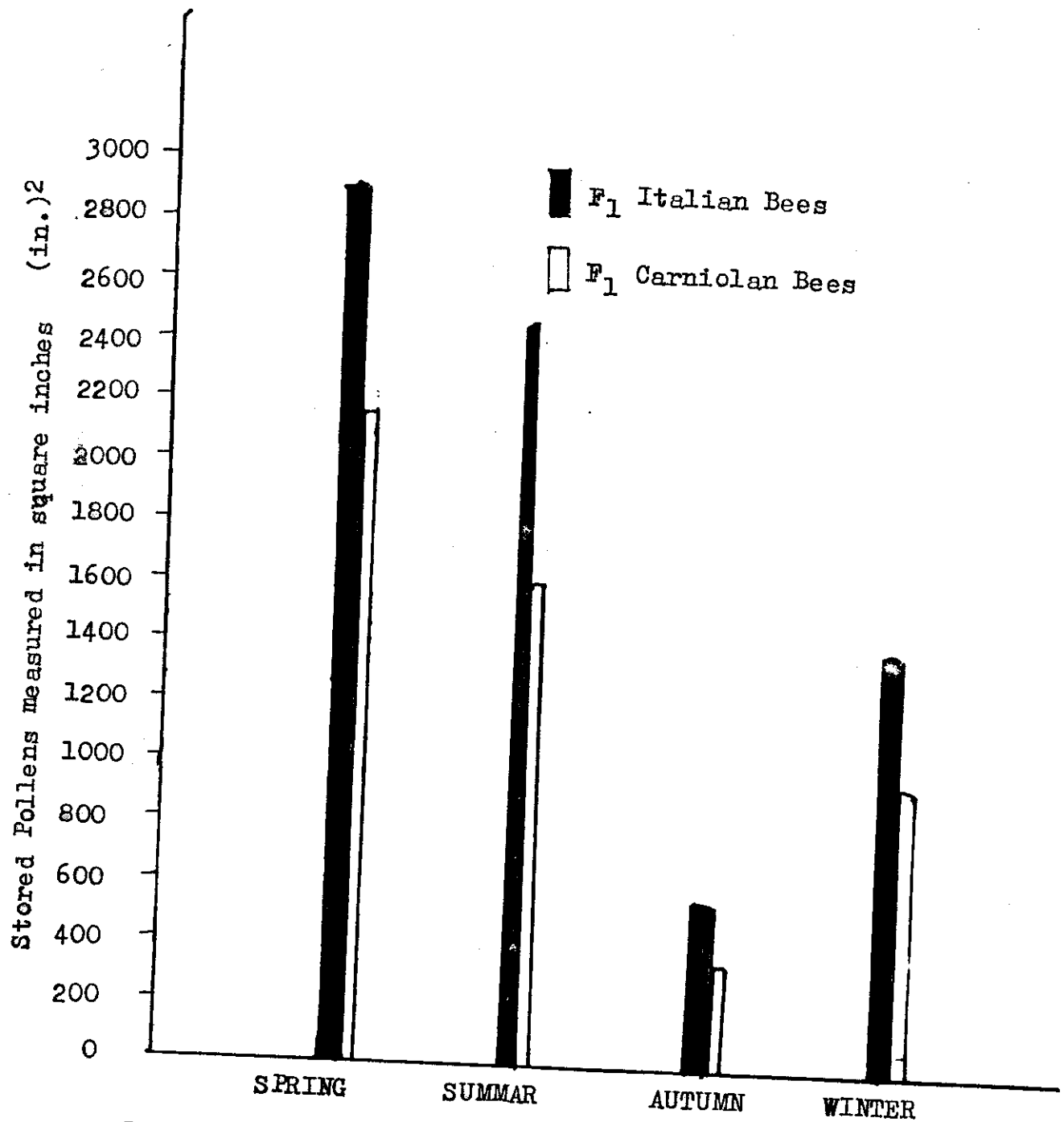


Fig. (24) The amounts of pollen stored in colonies during seasons (1986).

In Winter, F_1 Italian bees gathered 1413.74 in² and 9.29 g pollen and propolis respectively, while F_1 Carniolan bees collected 990.63 in² of pollen and 6.55 g of propolis. It was obvious that these amounts were more than the amounts collected in Autumn because on January and February there were broad bean and the blooming of deciduous trees.

It could be concluded that F_1 Italian bees are more suitable for rearing in Egypt as they produce more pollen, sealed brood and propolis than F_1 Carniolan bees.

This data agree with Attalah et al. (1988) who found that the total amount of pollen was greatest from Egyptian clover, followed by corn and broad bean.

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So, it is evident from table (22) that F_1 Italian bees gathered significantly more pollen than F_1 Carniolan bees under Al-Amar and Kaha regions.

3- Pollens Found in Propolis :

In this experiment samples of different propolis sources were examined under the microscope. The results recorded in Figs. (25 to 46) showed that , some types of pollens were clearly found in propolis extracts of different localities and seasons. Some of pollens were digested in propolis . The examination and photographs indicated that the colour of propolis, some components and volatile oils were founds.

These results may be in agreement with Snodgrass, 1956 , Ieyrish (1978) , Caillas (1978) and Konig (1985).



Fig. () Pollen found in Propolis at Honeybees
Research Center , Minist. of Agric. ,
El-Doky , Giza. (Mag. X₂₀₀).



Fig. () Pollen found in Propolis at Honeybees
Research Center , Minist. of Agric.,
El-Doky , Giza (Mag. X₂₀₀).

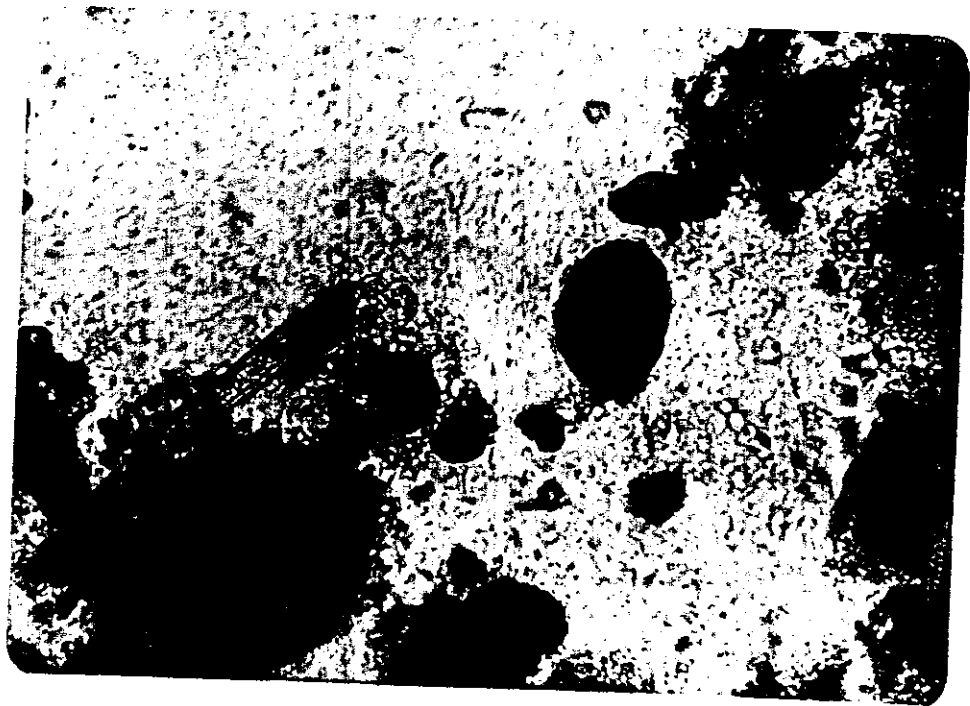


Fig. () Pollen found in Propolis at Honeybees
Research Center, Minist. of Agric. ,
El-Doky, Giza. (Mag. X₂₀₀)

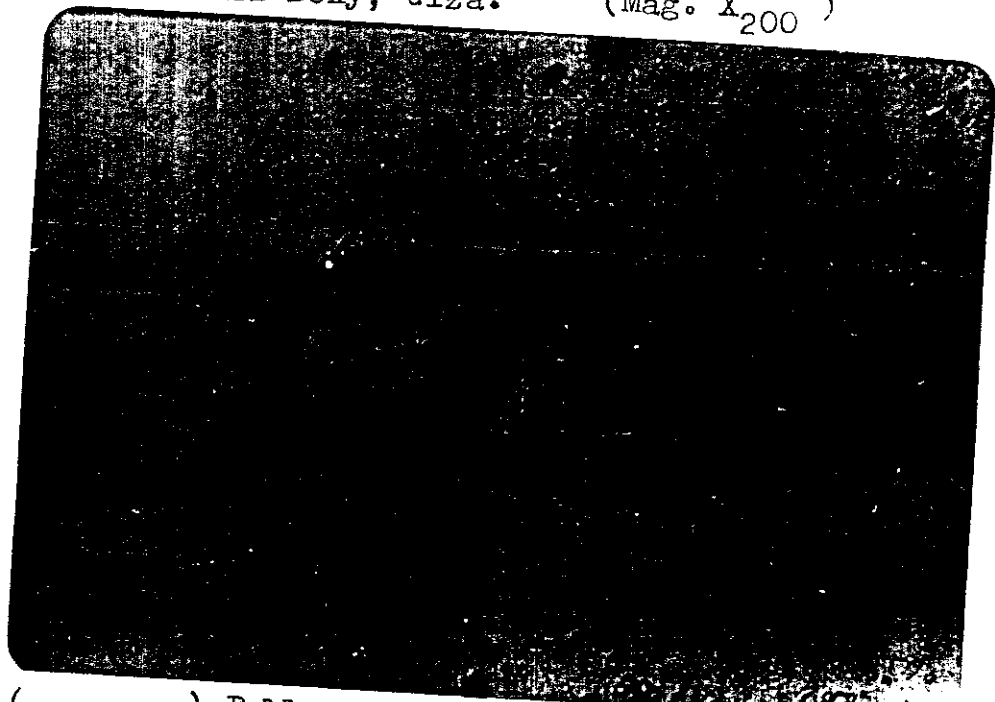


Fig. () Pollen found in Propolis at Honeybees
Research Center, Minist. of Agric. ,
El-Doky , Giza. (Mag. X₂₀₀).

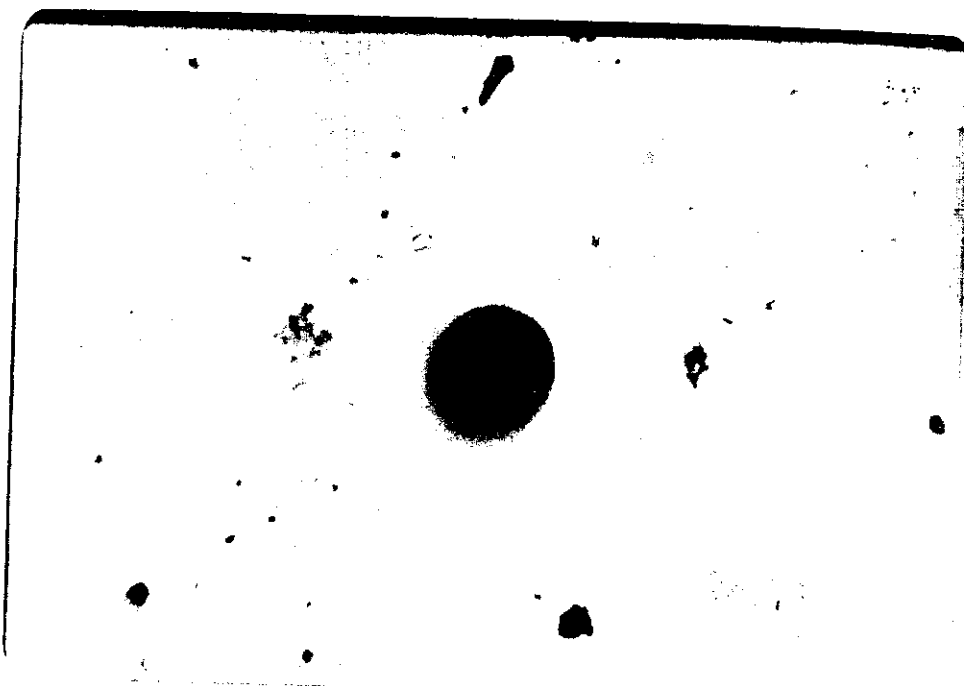


Fig. () Pollen found in Propolis from ashes
residue in soxhlet extract.
(Mag. X₂₀₀)



Fig.() Pollen found in Propolis from ashes
residue in soxhlet extract.
(Mag. X₂₀₀)



Fig. () Pollen found in Propolis gathered from inner clothes covers at Moshtohor region. (Mag. X_{200}).

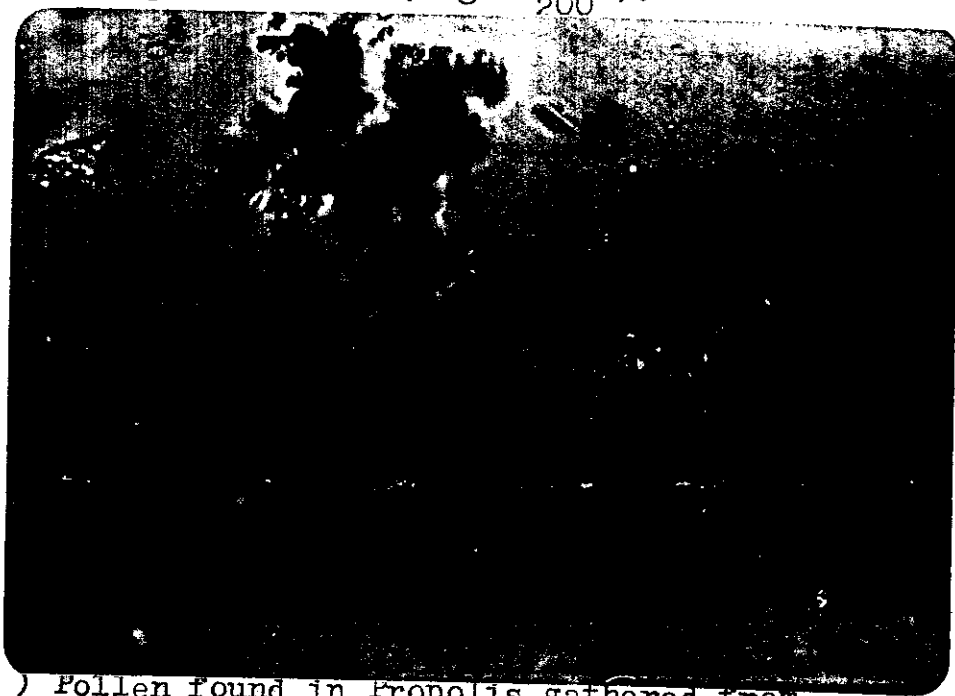


Fig. () Pollen found in Propolis gathered from inner clothes covers at Moshtohor region. (Mag. X_{200}) .



Fig. () Pollen found in Propolis (Alcohol extract)
from colonies keeping at Maha region.
(Mag. X_{200})

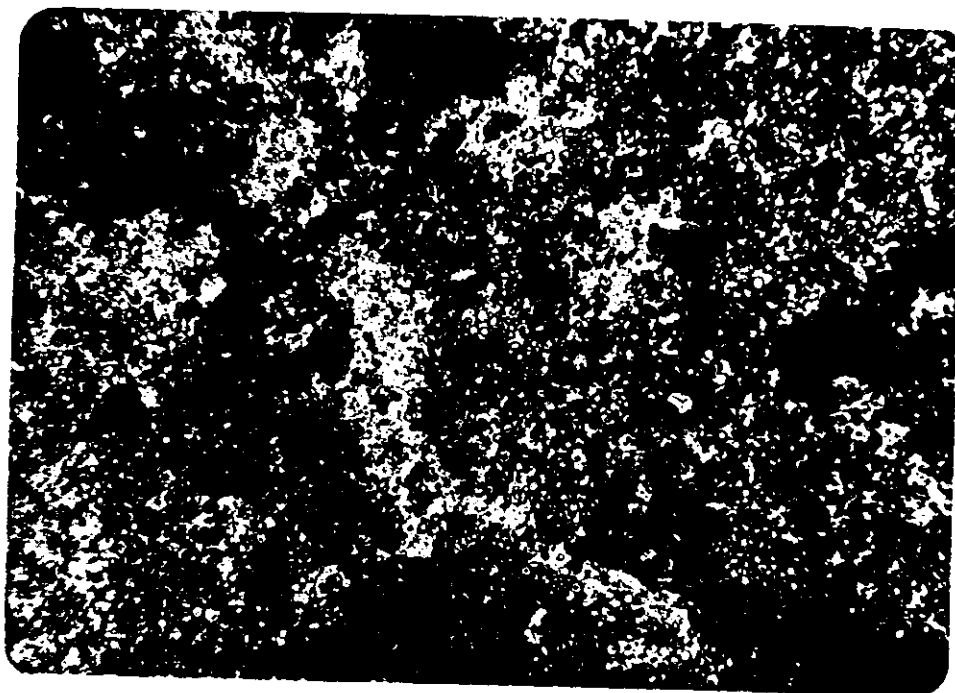


Fig. () Pollen found in propolis (Weeds pollen)
from colonies keeping at Maha region.
(Mag. X_{200}).



Fig. () Pollen found in Propolis (Alcohol extract).
(Moshtohor region) (Mag. X_{200})



Fig. () Some propolis components cristalized in
alcohol extracted. (Mag. X_{200}).

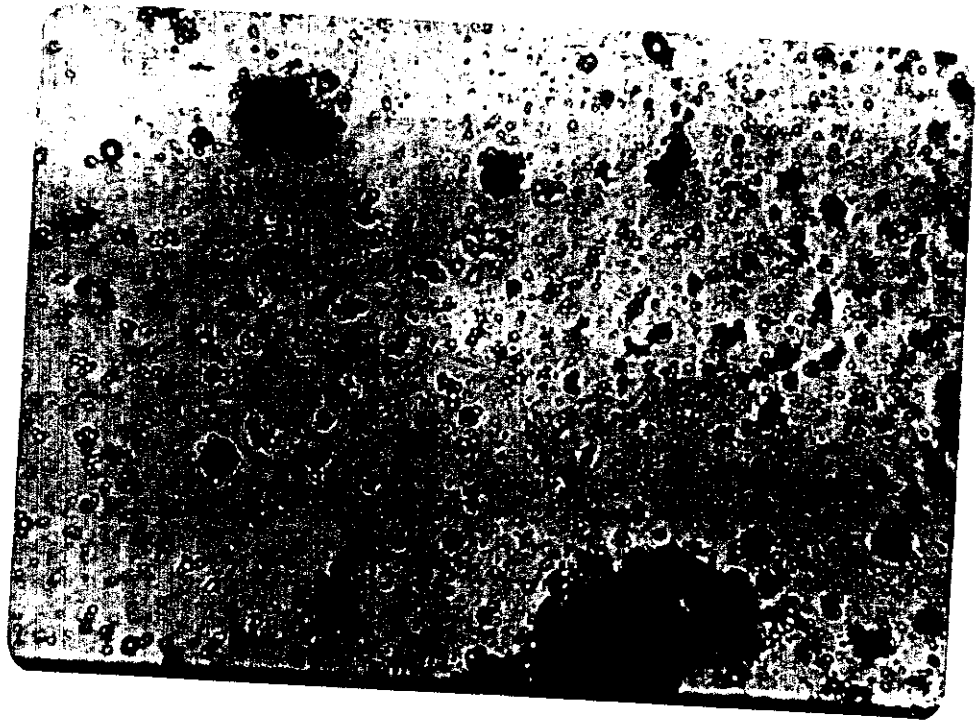


Fig. () Pollen found in Propolis (Alcohol extract).
(Mag. X_{200})



Fig. () Some components crystallized in Propolis
(Alcohol extract) (Mag. X_{200})



Fig. () Pollen found in Propolis (Alcohol-extract).

Mag. X 200)

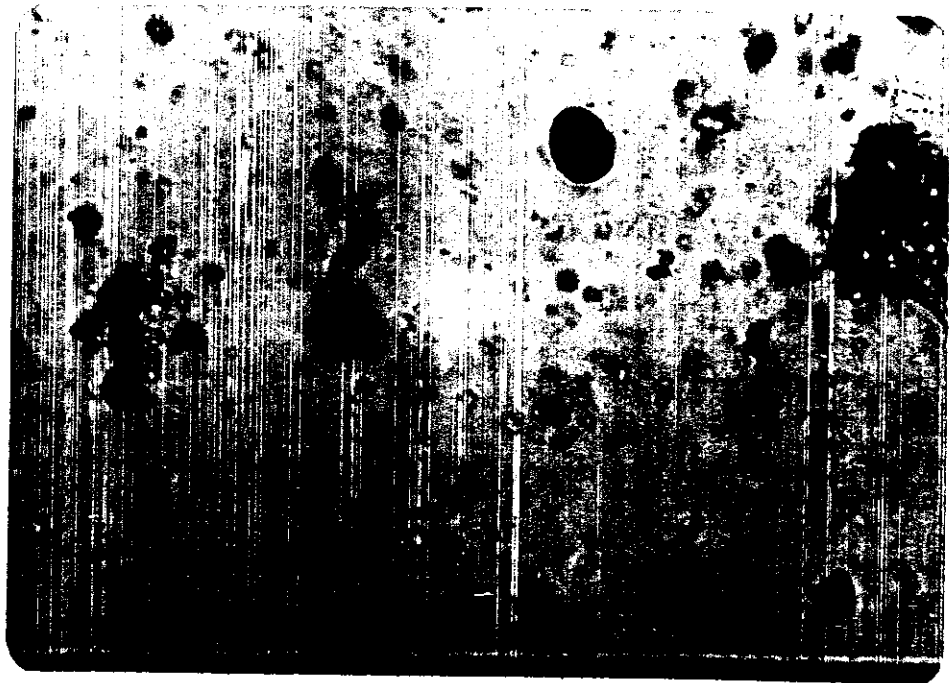


Fig. () Pollen found in Propolis (Alcohol extract).

(Mag. X 200)

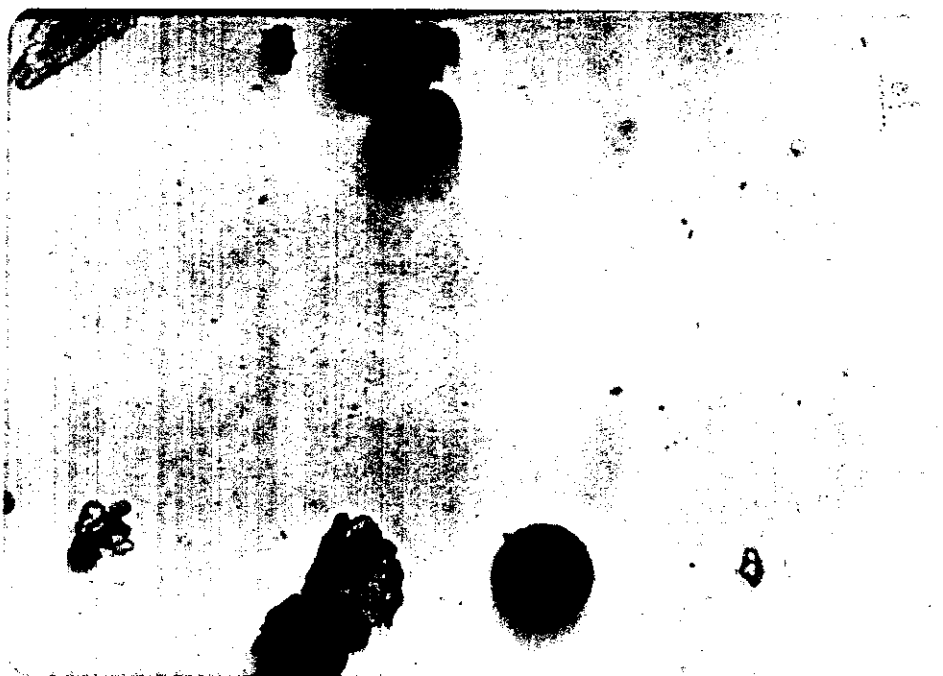


Fig. () Pollen found in Propolis (Alcohol extract).

(Mag. X₂₀₀)

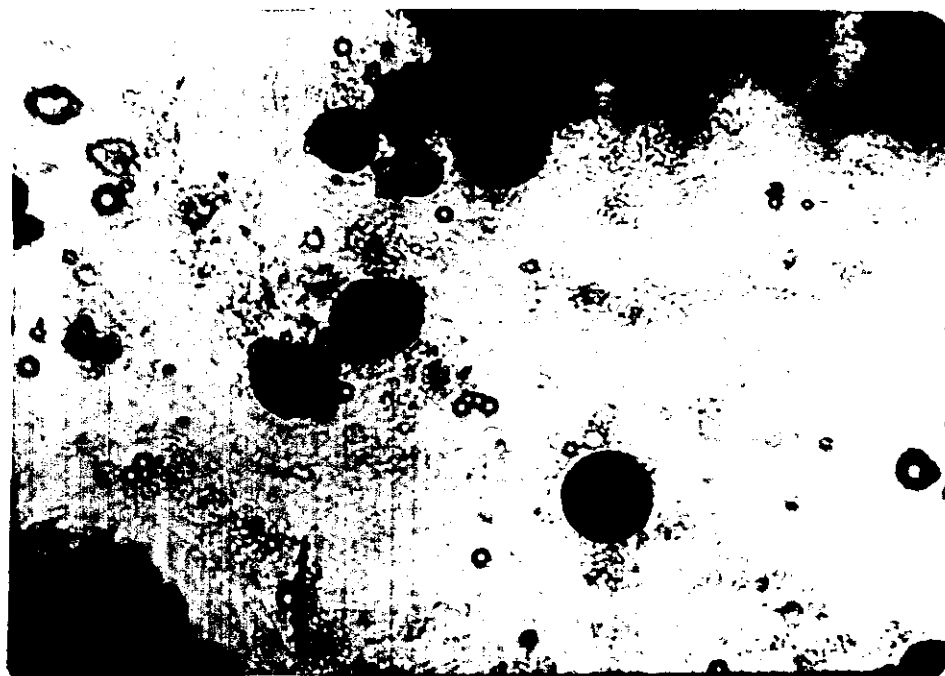


Fig. () Pollen found in Propolis (Alcohol extract).

(Mag. X₂₀₀)

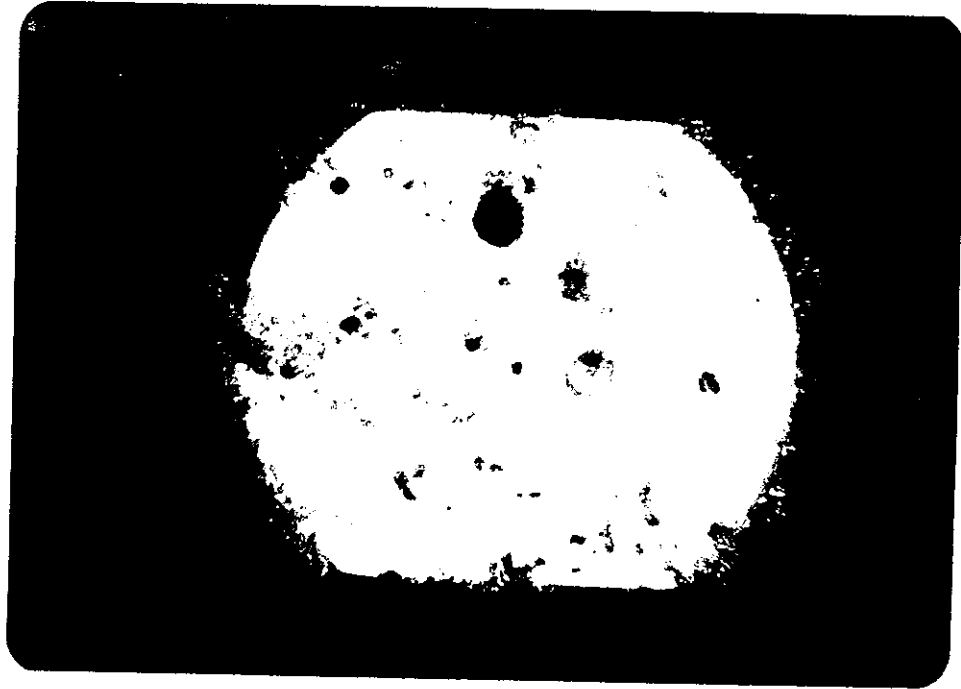


Fig. () Pollen found in Water propolis extracted
 (Some pollens digested)
 (Mag. X 100)

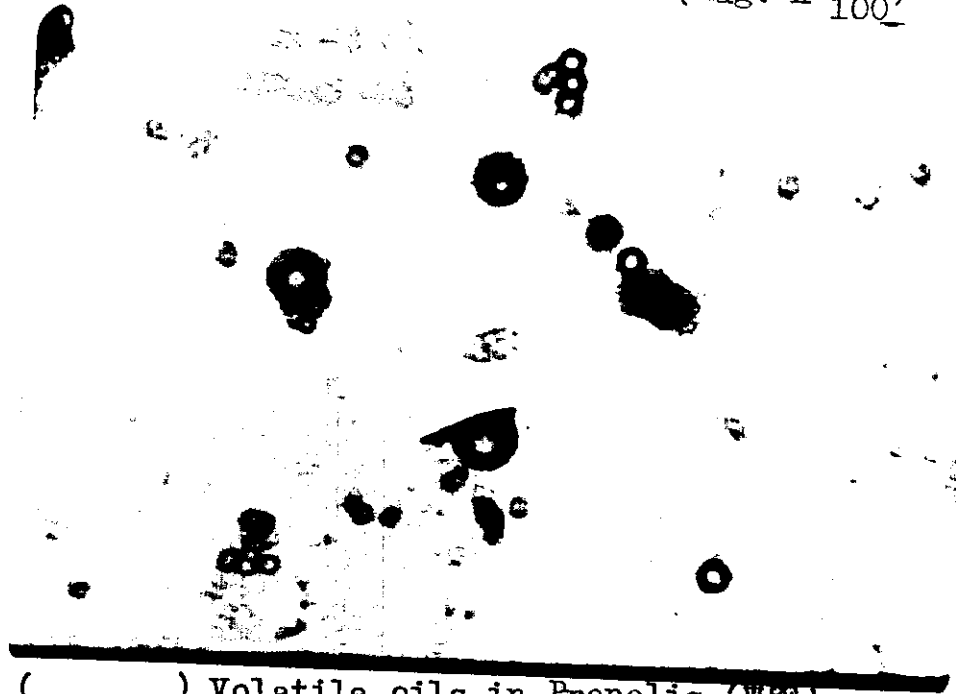


Fig. () Volatile oils in Propolis (WPE)
 (Mag. X₂₀₀)



Fig. () Pollen found in Water propolis extracted
(Mag. X₂₀₀)



Fig. () Pollen found in Water propolis extracted
(Mag. X₂₀₀).