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Streptococcal Disease
Among 93 families in Teheran, IRAN
A one - year study ***

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No previous study has been performed in Iran to show the prevalence of streptococcal infection and its late sequelae, namely rheumatic fever, rheumatic heart disease and glomerulonephritis,

Scattered information from pediatricians and physicians in town indicate that such diseases are quite prevalent. Also recent information from studies carried out in Thailand, Pakistan and Eastern Nigeria (1) show that contrary to what was previously believed, streptococcal infection and its sequelae are probably quite frequent in tropical and semi-tropical countries.

Therefore, to begin to obtain certain information with regard to streptococcal infection in Teheran, this study was undertaken to determine in a pilot experiment, and among two different socio-economic groups, the frequency of cases of pharyngitis, the prevalence of Streptococcus pyogenes in these cases , the carrier rate of beta hemolytic streptococci, the serologic groups and types of isolated strains and the incidence of rheumatic fever and glomerulonephritis, as observed in the period of study.

METHOD:

Selection of the families and design of study: In total, 446 people in 93 families in Teheran were included in this study, but 339 persons were regularly studied and followed during one full year.

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Admission to the study was limited to those who had children between the ages of 4 and 16 and the families were approximately equally divided into two different socio-economic groups called I and II. Roughly once every ten weeks, the families were visited at their homes by one of our technicians and throat cultures were taken from all the members and immediately inccubated on culture media. Throughout the year when a sore throat was reported, a technician would visit the patient and take a throat culture. Clinical signs and symptoms and results of throat culture were recorded on individual data sheets. To encourage of sore throat, free Calcipen * (300,000 I.U./3 times/ day for ten days) was given to individuals who had positive cultures.

MATERIALS:

Throat swabs were taken from patients or carriers and immediately inoculated on the surface of a 5 per cent defibrinated sheep blood agar plate. On arrival at the laboratory usually within half to one hour, the plates were subsequently streaked and incubated at 37 ° C for 18-24 hours. They were then examined for colonies of beta hemolytic streptococci and graded according to the yield into four groups.

less than 10 colonies *

Between 10 - 50 colonies **

Between 50 - 100 colonies ***

More than 100 colonies ****

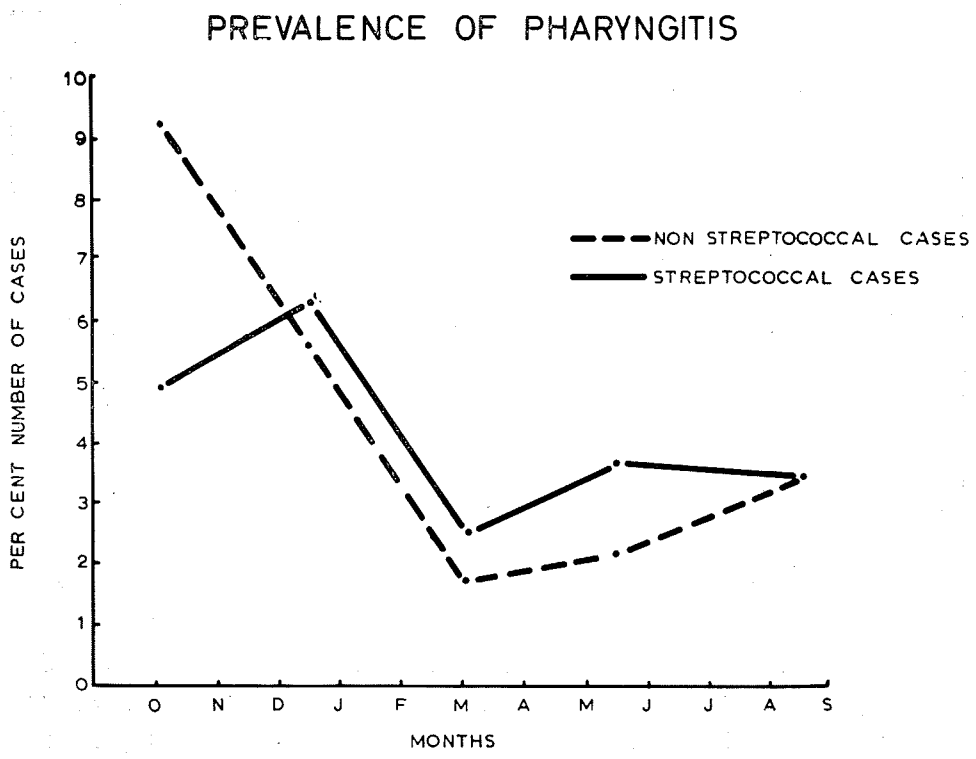
Representative colonies were subcultured and then grouped according to Lancefield's method (2, 3). Typing of group A streptococci was performed by Griffith's slide agglutination technique (4) . The specific grouping and typing sera against different serologic types were supplied by the WHO International Center for Streptococcus Typing, Institute of Epidemiology and Microbiology, Prague.

RESULTS :

During the twelve month period, there were 153 cases of pharyngitis. Of these, 72 (47.1%) patients had streptococcal and the remaining 81 (52.9%) had non-streptococcal pharyngitis. As seen in Figure I, the incidence of streptococcal sore throat in various months of the study period showed significant differences paralleling the fluctuation in overall pharyngitis. The highest incidence was from mid-October until the middle of December .

During the period of study definite rheumatic fever developed in a girl of seven and glomerulonephritis in a boy of eleven years old. This gives us a rate of 1.4 per cent each for rheumatic fever and glomerulonephritis following streptococcal sore throat.

The figure of 72 streptococcal sore throats among 339 people studied, gives us an overall infection rate of 21 per 100 persons per year.



Of the 72 (strains) isolated from patients, 35 (48.6.%) belonged to group A, 11 (15.2%) to group C, 4 (5.5%) to group G and unfortunantely 22 (30. 5%) were lost during subculturing and could not be grouped. Disregarding the 22 (strains) lost, the figures will be 70%, 22% and 8% for groups A, C and G respectively.

TABLE I: DISTRIBUTION OF DIFFERENT GROUPS OF BETA HEMOLYTIC STREPTOCOCCI ISOLATED FROM SORE THROAT CASES

STREPTOCOCCI GROUPS OF BETA HEMOLYTIC	1st. PERIOD OCT. 12-DEC. 21	2nd. PERIOD DEC. 22- FEB. 28	3rd. PERIOD MAR.1- MAY 18	4th. PERIOD MAY 19-JULY 25	5th. PERIOD JULY 26 - SEPT. 30	TOTAL	PER. CENT-AGE
A	14	9	4	5	3	35	48. 6%
B	-	-	-	-	-	-	-
C	5	1	3	-	-	11	15. 2%
G	-	-	-	4	4	4	5.5%
NOT GROUPED	12	3	4	1	2	22	30.5%
TOTAL OF ALL GROUPS						72	99.8%

From 153 cases of total pharyngitis 53 (34.6%) patients were over 16 and 100 (65. 2%) patients under. Similarly, among 72 streptococcal pharyngitis, 26 (36.1%) patients were over 16 and 46 (63.1%) patients under. From 35 cases of sore throat due to group A streptococci, 7 (20%) were among the over 16 and (80%) were among the under 16 age group.

There appeared to be a significant increase in the prevalence of total pharyngitis and pharyngitis due to all streptococci among the group under 16 years of age. This could be explained by the fact that older people through out the years have acquired more immunity to different specific M types of group A streptococci.

✿ Kindiy Supplied by Iran-pharmic Co. Teheran.

TABLE 11

AGE DISTRIBUTION OF PHARYNGITIS

AGE GROUPS	TOTAL CASES		PHARYNGITIS DUE TO BETA HEMOLYTIC STREPTOCOCCI		PHARYNGITIS DUE TO GROUP A STREPTOCOCCI	
	NO.	%	NO.	%	NO.	%
OVER 16	63	34.6	26	36.1	7	20
UNDER 16	100	65.2	46	63.9	28	80

TABLE III

SOCIO - ECONOMIC DISTRIBUTION OF PHARYNGITIS

SOCIO-ECONOMIC GROUPS	TOTAL CASES OF NONSTREP-TOCOCCAL PHARYNGITIS		PHARYNGITIS DUE TO BETA HEMOLYTIC STREPTOCOCCI		PHARYNGITIS DUE TO GROUP A BETA HEMOLYTIC STREPTOCOCCI	
	NO.	%	NO.	%	NO.	%
I	43	53.1	41	56.9	22	62.8
II	38	46.9	31	43.1	13	37.2

I - HIGH SOCIO - ECONOMIC GROUP

II - LOW SOCIO - ECONOMIC GROUP

There did not appear to be any significant difference in the prevalence of pharyngitis among the two different groups. The difference seen, particularly in the column of group A beta hemolytic streptococci, could be attributed to better facilities available to group I, such as the telephone which makes reporting easier.

	ALL AGE GROUPS			UNDER 16 YEARS			OVER 16 YEARS		
PERIODS	NUMBER	CARRIER	%	NUMBER	CARRIER	%	NUMBER	CARRIER	%
1st. PERIOD	446	60	13.4/	242	43	17.7%	204	17	8.3%
2nd. PERIOD	329	39	11.8%	197	25	12.6%	132	14	10.6%
3rd. PERIOD	354	52	14.6%	199	34	17.0%	155	18	11.6%
4th. PERIOD	333	35	10.5%	190	24	12.6%	143	11	7.6%
5th. PERIOD	234	37	15.8%	148	30	20.2%	86	7	8.1%
MEAN AVERAGE	339	44.6	13.2%	195	31.2	16,0%	144	13.4	9.2%

TABLE IV

CARRIER RATE OF BETA HEMOLYTIC STREPTOCOCCI AMONG 93 FAMILIES DURING ONE - YEAR STUDY

The overall carrier rate of 13.2 per cent obtained in this study is in agreement with other published reports (5,6).

There was some difference in this study between the carrier rate 16% among the under 16 year olds and the rate of 9.2% for those over 16 years of age.

There did not appear to be much difference between the carrier rates in two different socio- economic groups. Group I and II showed mean carrier rates of 12.2 and 14,1 percent respectively. Those under 16 had a carrier rate of 17.6 per cent in group I and 14.5 per cent in group II, but the exceptionally low rate of 6.4 per cent among those over 16 in group I could not be explained. As shown in table V, those over 16 in group II had 13.3 per cent carrier rate .

TABLE V

DISTRIBUTION AND COMPARISON OF CARRIER RATES OF BETA HEMOLYTIC STREPTOCOCCI BETWEEN TWO DIFFERENT SOCIOECONOMIC GROUPS

MEAN TOTAL OF ALL PERIODS	TOTAL NUMBER OF PEOPLE	NO. OF PEOPLE UNDER 16	NO. OF PEOPLE OVER 16	TOTAL NUMBER OF CARRIERS		NO. OF CARRIERS UNDER 16		NO. OF CARRIERS OVER 16	
				NO.	%	NO.	%	NO.	%
GROUP I	170	86	84	22	12.2	15	17.6	5	6.4
GROUPII	169	109	60	24	14.1	16	14.5	8	13.3

I HIGH SOCIO _ ECONOMIC GROUP

II LOW SOCIO - ECONOMIC GROUP

of 223 strains of beta hemolytic streptococci isolated from healthy carriers 96 (43%) belonged to group A, 6 (2.5%) to group B, 61 (27.3%) belonged to group C, 35 (15.6%) to group G, 6 (2,5%) were ungroupable 19 (8.6%) strains were lost during subculturing.

TABLE VI

DISTRIBUTION OF DIFFERENT GROUPS OF BETA HEMOLYTIC STREPTOCOCCI AMONG CARRIERS

GROUPS OF BETA HEMOLYTIC STREPTOCOCCI	1st. PERIOD	2nd. PERIOD	3rd. PERIOD	4th. PERIOD	5th. PERIOD	PERCENTAGE
A	21	17	15	24	19	43%
B	2	3	1	—	--	2 5%
C	19	14	11	8	9	27.3%
G	12	3	10	3	7	15.6%
LOST	4	2	13	--	--	8.6%
UNGROUPABLE	2	—	2	--	2	2.6%
TOTAL	60	29	52	35	37	99.5%

The colony count in urinary tract infection has proved itself quite useful and usually differentiates between infection and contamination. The same principle has been suggested in cases of pharyngitis as an index of streptococcal pathogenicity.

However, it can be seen from table VII that there was a great deal of overlapping between patients and carriers and therefore, the colony count failed to show a significant difference between carriers of beta homolytic streptococci and patients suffering from streptococcal sore throat.

TABLE VII

COMPARISON OF COLONIAL COUNTS OF BETA HEMOLYTIC STREPTOCOCCI BETWEEN PATIENTS AND CARRIERS

	LESS THAN 10 COLONIES		BETWEEN 10 - 50 COLONIES		BETWEEN 50 - 100 COLONIES		MORE THAN 100 COLONIES	
	NO.	%	NO.	%	NO.	%	NO.	%
PATIENTS	9	13.6	3	4.5	22	33.3	32	48.4
CARRIERS	72	28.1	52	20.2	99	38.2	34	13.2

As seen in table VIII, it was possible to type all group A strains by agglutination (Ttyping). 77 per cent of group A strains isolated from patients were included in two prevalent tyoes, namely types 5/11/12/27/44 (48.5%) and 3/13/B 32$_6$4 (28,5%). Among the carriers the most prevalent types were type 1 (26%), type 3/13/B 3264 (13.5%) type 2 (12.5%0 and type 5/11/12/ 27/44 (19.8%).

TABLE VIII

DISTRIBUTIN OF TYPES* OF S. PYOGENES AMONG PATIENTS AND CARRIERS

TYPE	PATIENTS		CARRIERS	
	NO.	PERCEN-TAGE	NO.	PERCENTAGE
1	2	5.7	25	26
2	1	2.9	12	12.5
3/13/B 3264	10	28.5	13	13.5
4	0	0	5	5.2
5/11/12/27/44	17	48.5	19	19.8
6	0	0	4	4.1
8/25/IM p 19	0	0	5	5 2
9	1	2.9	1	1.1
14	2	5.7	5	5.2
15/17/19/23/30/47	2	5.7	5	5.2
28	0	0	2	2.1
TOTAL	35	99.9	96	99.9

* Tayglutinating antigen

DISCUSSION:

This presentation, being a pilot study, the data in it are restricted and the figures obtained are in many ways too small to permit a definite opinion on the magnitude of streptococcal infection and its late sequelae. It was also unfortunate that due to the strong refusal of families concerned, no immunological studies of streptococcal infection could be performed. However, some of the figures in this study warrant further detailed work, e.g. our figure of 21 for an overall infection rate for 100 persons per year is at the upper limit of some of the similar types of study undertaken elaswhere (7,8,9,)

It is also suggestcd that between 25 to 60 par cent of all clinical illnesses due to sore throat are of streptococcal etiology, and the proportion of group A streptococcal varies from 70 to 90% in cases of pharyngitis and upper respiratory infection to 60 to 84% in carriers (9, 10). In the present study 47 per cent of the sore throats werc of stroptococcal origin, but the proportion of groups A strains among patients was 486 per cent and among carriers 43 per cent. Therefore, in our study. group A prevalence was not as high, and further , group C and G apeared to play a more important role in pharyngitis than they usually appear to do in studies of this nature

Similar finding i.e.higher incidence of sore throats due to groups C and G were indicated by Dr. Padmavali from India and Dr. Abdin from Egypt at the Eighth Intenational Congresses of Tropical Medicine and Malaria in September 1968 in Teheran.

It wnuld be of intrest to know whether groups C and G strains of beta hemolytic streptococci in tropical and semi – tropical areas show a greater pathogenicity.

The fact that 94% of all strains isolated from patients were included in fi e prevalent types emphasizes the importance of type identification if the possibility of prevention of infection by use of vaccines becomes a reality.

SUMMARY

During a one year period of investigation in Teheran an average of 339 people in 93 families and from two different socio-economic levels wesre regularly studied and followed for streptococcal infection.

A significant increase in both streptococcal and non - streptococcal pharyngitis was seen during October and November. of 153 cases of phar yngitis seen, 53 (34.6%) patients were over 16 and 100 (65.2%) were under 16. 47 per cent of patients harbored beta hemolytic streptococcal, hence the infection rate per 100 persons per year for streptococcal sore throat was 21.2 per cent. No significant difference was secn with regard to pharyng-itis among the two socio-economic groups. The overall carrier rate for beta hemolytic streptoc-occi during the year was 13.2%. The rate for the under 16 years was 16.0% and 9.2% for the over 16' Group I had a carrier rate of 12.2% and Group II, 14.1%. Virtually all group A strains were typable and those isolated from patients belonged to five prevalent types.

During the period of study, definite rheumatic fever developed in a girl of seven and glomerulonephritis in a boy of eleven years old.

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TABLE III

NUMBER OF MICE	IRRADIATION DOSE	DAYS AFTER IRRADIATION	WHITE CELLS COUNTS	PLATELETS COUNT	VARIATION OF THE PER CENT OF THE WHITE CELLS	VARIATION OF THE PER CENT OF THE PLATELETS
3a	—	0	16600	504000	—	—
3a	112 R	3	12400	432000	- 25	- 14.2
3a	112 R	7	12900	448000	- 23	- 11
3a	112 R	10	12800	496000	- 23	- 1.6
3b	—	0	16000	432000	—	—
3b	112 R	3	12100	376000	- 24	- 13
3b	112 R	7	12300	424000	- 22	- 1.8
3b	112 R	10	—	472000	- 23	9
3c	—	0	17600	512000	—	—
3c	112 R	3	13400	432000	- 23.7	- 14
3c	112 R	13	16200	480000	- 4.5	- 6.2

Table III shows clearly that :

1- The maximum reduction of the white cells and platelets is on the 3rd day after irradiation.

2- The amount of reduction of white cells and platelets is nearly constant ; for the white cells it is 23% and for the platelets is 13%. These two figures were chosen in the following experiments for the comparison of the degree of protection given by Iranian Manna.

II. In 4 mice before and on the 3rd day after injection of Iranian Manna and irradiation, the white cells, and platelets were calculated. Comparing their reductions with Table III (23% reduction for white cells, and 13% reduction for platelets), the degree of protection by manna was determined. This is shown in Table IV.

TABLE IV

Number of Mice	Quantity of the Protector	Dose	Hours after Irradiation	White Cells Counts	Platelets Counts	Variation of the % of the white cells cells	Variation of the % of the platelets	Degree of the protection for white cells with comparison to the 23% of the Table III	Degree of protection for platelets with comparison to the 13% from Table II
1a	5cc of Iran ian Manna of saturated solution frcm 5cc diluted in water	—	—	16700	496000	—	—	—	—
1a		112R	72	12400	480000	- 26 %	- 3 %	0 %	79 %
2a	»	—	—	18200	667000	—	—	—	—
2a		112R	75	14800	648000	- 18 %	- 3 %	25 %	79 %
3a	»	—	—	16100	488000	—	—	—	—
3a		112R	72	10900	328000	- 32 %	- 32 %	0 %	0 %
4a	»	—	—	18200	360000	—	—	—	—
4a		112R	72	14100	352000	- 22 %	- 2 %	9 %	72 %

TABLE V

Number of Mice	Quantity of the Protector Injected before Irradiation	Irradiation Dose	Hours after Irradiation	White Cells Counts	Platelets Counts	Variation of the % of the white cells	Variation of the % of the white cells	Degree of protection for white cells with comparison of 23% of Table III	Degree of protection for Platelets with comparison of 13% of Table III
5 a	1/10 mgr. of KCN	-	-	17400	53600	-	-	-	-
5 a		112R	72	12300	512000	- 29 %	- 4.4 %	0 %	69 %
6 a	»	-	-	11800	552000	-	-	-	—
7 a	.05 mgr of KCN	-	-	13800	536000	-	-	-	-
7 a		112R	72	10200	448000	-26.5%	- 16.4 %	0 %	0 %
8 a	,066 mgr. of KCN	-	-	16500	488000	-	-	-	-
8 a		112R	72	11300	408000	31. 5%	- 16%	0 %	0 %
9 a	.075 mgr. of KCN	-	-	26700	504000	-	-	-	-
9 a		112R	72	21000	488000	- 21.3%	3.1 %	12 %	78 %
10a	»	-	-	26100	480000	—	-	-	-
10a	»	112R	72	19600	456000	- 24.9 %	- 5 %	0 %	64.3%

* Because of the more poisoning of the KCN than the NaCN we used less than 1/10 miligram of potassium cyanide to prevent the death of the mice as shown in the above table.

Table IV: Indicates a comparison of % reduction of Platelets and white cells in the mouse irradiated and injected, and the irradiated but not injectod.

From Table IV the result is:

The rcduction of white cells and platelets in the injected mice is 63% and 20% as compared with mice not previously injected.

III. In another series of experiments which have been done in several mice, the effect of the protection by Iranian Manna and potassium cyanide were compared. Table V shows the results of KCC protection degree.

RESULTS :

The comparison of Table V with Table IV is shown below :

1- The protective effect of Iranian Manna on the platelets is higher than with KCN and the percentages of protection in 3 out of 4mice are found to be 79% and 72%. Comparing the potassium cyanide the effect of protection appeared in 3 out of 5 mice and the percentages rewe 69% , 78% and 64. 3%

2- For the white cells the effect of Iranian Manna as a protector in 2 out of 4 mice was 25% and 9%, while in potassium cyanide only in one mouse out of 5 the effect of the protector appeared and the percentage of it was 12 %.

CONCLUSION :

From all of the above experiments it is proven that Iranian Manna is one of the chemical protectors against ionizing radiations. Regarding poisoning, it was shown that it contains no poison at all. Large doses through injections or through eating can be used because of the absence toxicity. The mechanism of the effect of Iranian Manna protection is perhaps due to the presence of the reducing agents of glucoside which is contained in Iranian Manna. Perhaps some other elements are contained in Iranian Manna which are responsible for its being protective. The existence of the plant in arid desert regions might account for its developing a defence against radiation. It appears to be advisable to do further resarch on the formula of Iranian Manna.

SUMMARY

1 - The Iranian Manna as a Protector against Ionizing Radiation reduces :

A. The increase of gammaglobulin in mice caused by x -- ray radiation.

B. Rate of death in mice caused by x - ray radiation.

C. 1. The appearance of vacuoles in the red cells of the pigeon caused by ionizing radiation.

2. The decrease of the Platelets and white cells in the pigeon caused by ionizing radiation.

2- A. Determination of the percentage of the protective effect of the Iranian Manna by comparison of irradiated with non - irradiated mice .

B The comparison of the protective effect of Iranian Manna with that of Potassium Cyanide.

SOMMAIRE

I. La manne de perse (Atrophaxis Spinosa), employee comme un protecleur de radiations ionisantes reduit:

1- la diminution de gammaglobuline de sang des souris causée apr les rayons x.

2- la mortalité survenue par les rayons x.

3- làpparition des vacuoles dans les globules rouges des pigeons causée par les radiations ionisiantes.

II. a. La determination du pourcentage de l'effet protecteur de la Manne de Perse par la comparison entre les souris irradiées et non irradiées sur globules blancs et en Plaquettes.

b. la comparsison de l'action protectrice de la Manna de Perse avec celle du cyanure de potassium.

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