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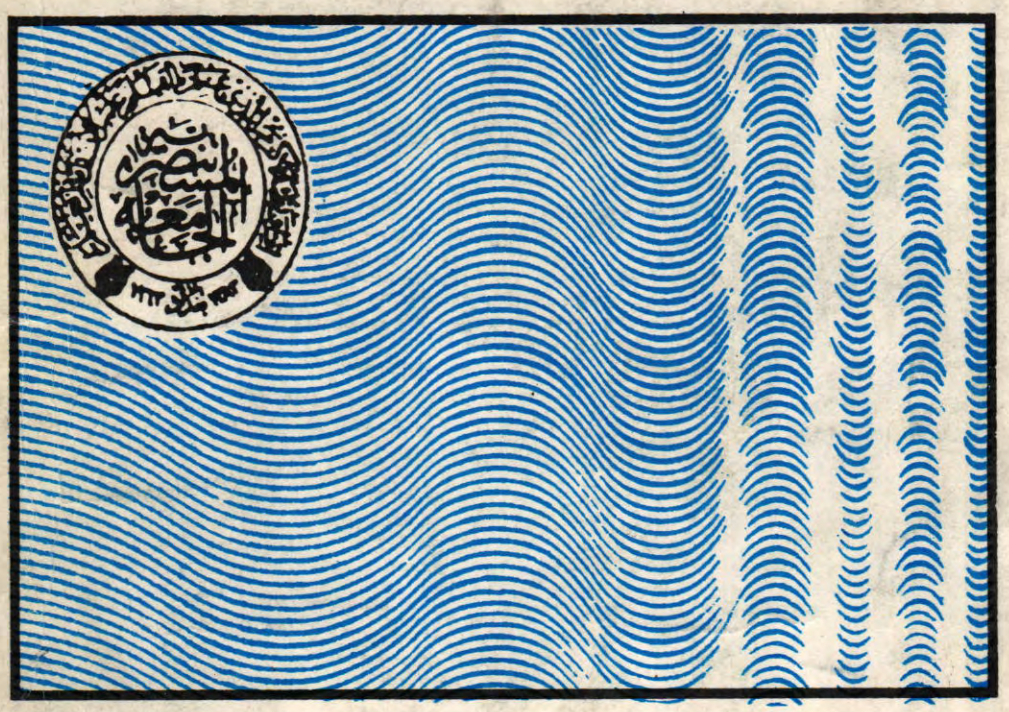

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المحتويات

البحوث الانكليزية

Page No.

A Comparative Study of Media for The Isolation of Salmonellae and Shigellae from Diarrheal Stool Samples

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الخلاصة

قورنت كفاءة ثلاثة اوساط الثنائية وستة اوساط تنمية في عزل بكتريا السالمونيلا والسغيلا من (٦٨٠) عينة خروج من مرضى اربع مستشفيات في بغداد . أظهرت النتائج ان اغناء العينات بوسط التتراثايونيت (Tt) ومن ثم نقلها الى وسط التنمية الهكتون (HE) كانت اكفا طرائق عزل السالمونيلا (٧٩,١٪) تليها طريقة استخدام السلفايت (SF) كوسط اغنائي والبزموث اكار (BSA) كوسط تنمية وبنسبة عزل بلغت (٧٧,٧٪) ، ولكن الطريقة الاخيرة كانت اكثر كفاءة في عزل النوع *S. typhi* . كما لوحظ ان افضل طريقة لعزل الشغلات من عينات الخروج هي باستخدام وسط السلفايت الاغنائي منالهكتون او وسط الكرام السالب (GN) مع اكار الديسوكسي كوليت (DCA) اذ اعطت كلا من هاتين الطريقتين كفاءة عزل بلغت (٦١,٩٪) تليها طريقة استخدام السلفايت مع اكار الديسوكسي كوليت وبنسبة (٥٩,١٪) . لم تثبت هذه الدراسة كفاءة وسط السالونيلا والشغيلا (SS) الذي يستخدم عادة في مختبرات المستشفيات المحلية عند عزل السالمونيلا والشغيلا من خروج المرضى .

ABSTRACT

Efficiency of three enrichment broths , "Tetrachionate (Tt) , Selenite-Facese (SF) and Gram Negative (GN)" , and six plating media "Hekton Enteric (HE) Agar , Bismuth Agar (BA) , Brilliant Green Agar (BGA) , Desoxycholate Citrate Aga (DCA) , Salmonella-Shigella (SS) Agar and MacConky Agar (Mac)" for the isolation of Salmonellae and Shigellae from (680) diarrheal stool samples of hospitalized patients were studied and compared. Using Tt as an enrichment broth with HE agar as a plating medium was the most efficient method for the isolation of Salmonellae with recovery rate of 79.1% ; followed by SF broth with Bismuth Sulfite Agar (BSA) at a rate of 77.7%. However , the later combination proved to be better in isolating *S. typhi* . For the isolation of Shigellae , GN broth with DCA and SF with HE were the best combinations at the rate of 61.9% ; followed by SF with DCA (59.1%) . Unfortunately , SS agar which is used by most of the local clinical laboratories for the isolation of both Salmonellae and Shigellae did not prove to be that efficient for the recovery of the organisms under study.

INTRODUCTION

Traditional methodology for the detection of enteric pathogens invariably employs a combination of two media which includes an enrichment broth and a differential plating agar (1) .

Dependability in recovery of pathogens such as Salmonellae and Shigellae from faecal or non faecal samples is a serious problem for all bacteriology laboratories (2).

Previously , several methods for the isolation of enteric pathogens from stool were suggested ; most of these methods used enrichment media (i.e Tt and SF) as the common frequent media for the isolation of Salmonella (3) . A selective enrichment medium was described by Hajna (1955) for gram negative bacilli and he noticed increasing in the isolation rate for Shigellae using this medium in comparison with the direct methods (4) .

Enrichment media should be subcultured on a selective solid medium . in order to isolate Shigellae, a medium of a relatively low selectivity (eg. DCA) should be used . If Salmonellae needed to be isolated , then a more selective medium has to be used "i.e using Bismuth in the case of *S. typhi*"(5).

In attempt to achieve an optimal method (with high recovery) for isolating suspected Salmonellae and Shigellae from hospitalized patients stools , different enrichment broths and plating media were evaluated.

MATERIALS AND METHODS

Source of Samples

Diarrheal stool samples were collected from 680 patients of different ages at four hospitals in Baghdad ; two were for children hospitals and the others were general . Half of the samples were taken during winter (Dec. 1991 - Feb. 1992) and the other half during summer (June - Aug. 1992) .

Culture Media

A. Enrichment broths

Three enrichment broth namely ; Gram Negative (GN) broth (Di fco) , Tetrathionate (Tt) broth and Selenite - Faeces (SF) broth (Oxoid) were used . Tt and SF were boiled in a water bath for 10 minutes. While GN sterilized in the autoclave at 121 °C for 15 minutes . Two drops of iodine solution (55%) were added to each 10 ml. of sterilized Tetrathionate broth before inoculation (6).

B. Plating media

Bismuth Sulfite Agar (BSA), Desoxycholate Citrate Agar (DCA) , MacConkey (Mac) agar and Salmonellae - Shigellae (SS) agar by Oxoid ; Hekton enteric (HE) agar by Biomeriux ; Brilliant Green Agar (BGA) prepared from its constituents (6) . BSA , DCA , DCA, HE and SS were boiled for one minute . BGA and Mac were sterilized in the autoclave at 121°C for 15 minutes.

Method of Testing

Ten ml. of each three enrichment broths were inoculated with approximately one ml. of diarrhea stool sample , then hand shaked for homogenization . Emulsions were incubated for 24 hrs. at 37 °C , then streaked onto plates of solid medium . Plates were incubated for 24 hrs. at 37 °C except BGA and BSA for 48 hrs. Colonies suspected to be Salmonellae or Shigellae were identified biochemically (6) and serologically with welcome antisera , The final serological analysis was confirmed by the National Salmonellae Center in Baghdad .

RESULTS

Salmonellae recovery

A total of 67 strains of Salmonellae were isolated from the 680 stool samples tested . Numbers and percentages of Salmonellae isolates by each method were shown in table 1 .

The highest mean recovery rate (57%) was obtained by using Tt as an enrichment broth , followed by SF (52.2%) and GN (35.8%) which is

considered to be insufficient enrichment broth for Salmonellae .

For plating media , HE agar resulted in the highest recovery for isolation of Salmonellae from stool with 66.7% as average percentage for the three enrichment broths ; followed by BSA (58.7%) and BGA (54.7%) . The efficiency of SS medium was not proven , in this study , to be an efficient medium when its recovery rate was 38.8% .

Table 1 shows that the best method for the isolation of Salmonellae from stool is by using Tt as enrichment broth then streaking on HE agar as a plating medium with an isolating rate of 79.1% , followed by SF with BSA at a rate of 77.6% . However , later method was superior in recovery of *S. typhi* , there was inconsiderable relationship between the type of Salmonellae and the isolating method.

Shigellae recovery

Table 3 shows the distribution of Shigellae isolates recovered by enrichment and plating media studied . Enrichment media SF and GN were efficient in increasing the frequency of recovery up to 50% and 48.8% , respectively . In contrast , Tt resulted in lower recovery rate (27.4%) .

Table 1 : Distribution of the 67 isolates of Salmonellae recoverd by various enrichment and plating media

Enrichment broths / Plating media	Tt		SF		GN		Total	
	No.	%	No.	%	No.	%	No.	%
HE	53(67)	79.1	48(67)	71.6	33(67)	49.3	134(201)	66.7
BSA	39(67)	58.2	52(67)	77.6	27(67)	40.3	118(201)	58.7
BGA	50(67)	74.6	32(67)	47.8	28(67)	41.8	110(201)	54.7
DCA	37(67)	55.2	27(67)	40.3	22(67)	32.8	86(201)	42.8
SS	29(67)	43.3	31(67)	46.3	18(67)	26.9	78(201)	38.8
Mac	21(67)	41.3	20(67)	29.9	16(67)	23.9	57(201)	28.4
Total	229(402)	57.0	210(402)	52.2	144(402)	35.8		

Table 2: Salmonella Species recovered by various isolation methods

Salmonella Sp.	No	HE	BSA	BGA	DCA	SS	Mac
		Tt SF GN	Tt SF GN	Tt SF GN	Tt SF GN	Tt SF GN	Tt SF GN
S.give	31	26 23 16	15 23 13	24 17 13	18 10 9	11 12 9	8 8 6
S.aona	12	11 11 7	8 10 4	10 5 7	8 5 5	6 6 4	5 2 3
S.typhivnurium	10	8 6 6	6 8 5	7 6 6	7 6 6	7 7 3	4 6 4
S.seftenberg	3	3 3 2	3 2 2	3 1 1	2 1 0	1 0 1	2 1 0
S.worthington	3	2 3 1	1 2 1	2 1 0	1 1 0	1 2 1	1 1 1
S.enteritidis	3	1 1 1	2 3 1	2 1 1	0 2 1	2 1 0	0 1 1
S.typhi	3	0 1 0	2 3 1	0 0 0	0 1 0	1 2 0	0 1 0
S.paratyphi B	2	2 0 0	2 1 0	2 0 0	1 1 1	0 1 0	1 0 1
Total	67	53 48 33	39 52 27	50 32 28	37 27 22	29 31 18	21 20 16

Results of this study confirmed that DCA was the best plating medium for isolation Shigllae with a total average for the three enrichment broths (52.4%), followed by HE (46.6%) . MacConkey agar was better than SS agar .

Both methods of isolation ; GN with DCA and SF with HE , gave a recovery rate of 61.9% . Followed by SF with DCA with a rate of 57.7%. No correlation was found between isolating method and Shigella SPP recovery (Table 4) . In general , *S. flexneri* and *S. sonni* were more capable to grow on the used media than *S. dysenterae*.

DISCUSSION

Since it is impractical to investigate all types of media used for the isolation of enteric pathogens from clinical specimen , only those available in clinical bacteriological laboratories were evaluated in this study .

Most of the enrichment media are highly selective , and their selectivity are based on ability to support the growth of enteric pathogens as well as to inhibit other intestinal organisms .

In our study , Salmonellae are isolated most frequently after enrichment with Tetrathionate . Similar results were found by Dunn and Martin (2) when they noticed that Tt gave a recovery rate at 75.6% as a compared with 62.5% and 54.8% for SF and GN , respectively . For the isolation of Salmonellae from food Tt is better than SF (7.8). Selection of an enrichment medium for the isolation of Salmonellae depends on the number of the competing bacteria . As long as coliforms usually present in large numbers in the stool samples and sewagematerial , Tt enrichment broth is of higher value than SF (9).

Table 3 : Distribution of the 21 isolates of Shigellae recovered by various enrichment and plating media.

Enrichment broth / Plating media	Tt		SF		GN		Total	
	No.	%	No.	%	No.	%	No.	%
HE	6(21)	28.6	13(21)	61.9	11(21)	52.4	30(63)	47.6
DCA	8(21)	38.1	12(21)	57.1	13(21)	61.9	33(63)	52.4
SS	5(21)	23.8	7(21)	33.4	9(21)	42.8	21(63)	33.3
Mac	4(21)	19.0	10(21)	47.6	8(21)	31.2	22(63)	34.9
Total	23(84)	27.4	42(84)	50.0	41(84)	48.8		

Table 4: Shigella species recovered by various isolation methods.

Type of Salmonellae	No.	HE			DCA			SS			Mac		
		Tt	SF	GN	Tt	SF	GN	Tt	SF	GN	Tt	SF	GN
S. flexneri	12	4	8	7	5	6	7	3	4	4	2	4	3
S.sonnei	4	2	3	2	2	2	3	1	2	3	2	3	2
S.dysenteriae	4	0	2	1	0	3	3	1	1	1	0	2	2
S.boydii	1	0	0	1	1	1	0	0	0	1	0	1	1
Total	21	6	13	11	8	12	13	5	7	9	4	10	8

Shingella is a fastidious organism which can not reduce Tetrathionate (10) , thus it fails to grow in such broth . Instead of that SF and GN are usually the media of choice . Present results agree with those found by Dunn and Martin (2) when they noticed the SF and GN broths were found to be better than Tt in recovering Shegellae.

The plating media were chosen from a stand point that the categories of "selective" and "highly selective" . The results show HE agar was superior plating medium for isolating both Salmonellae

and Shigellae . This was fairly close to that mentioned by King and Matzgar (11) when they found HE superior to SS in the isolation of such enteric pathogens . Moreover , BSA and BGA were good plating media for the isolation of Salmonellae when compared with SS because of the presence of brilliant green dye.

Shigellae is known to be more sensitive to the extrinsic environment than Salmonellae , thus MacConkey is preferred for isolating Shigellae than SS . This was noticed by Morris to SS in isolating Shigellae from stool due to the citrate and thiosulphate salts contents of the latter.

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The Use of Electrophoretic Soluble Proteins Patterns for Grouping of *Rhizobium* and *Agrobacterium*.

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الخلاصة

اعتمدت مخططات الهجرة الكهربائية للبروتينات الذائبة بأستخدام هلام البولي اكريلمايد للتعرف بشكل مجاميع على سلالات مختلفة من بكتريا *Rhizbium* والـ *Agrobacterium* أظهرت نتائج البحث بعدم تجانس السلالات ، غالبية سلالات *R.meliloti* وكذلك *R.trifolii* وجدت متجانسة ، في حين ظهرت اختلافات بين سلالات *R.leguminosarum* و *Agrobacterium* . تقترح النتائج بانه يمكن الاعتماد على مخططات الهجرة الكهربائية للبروتينات الذائبة لهذه السلالات في التصنيف وانها مطابقة الى نتائج سابقة حصلت عليها باعتماد طريقة التصنيف العددي .

ABSTRACT

Soluble protein patterns obtained by polyacrylamide gel electrophoresis was used to group different strains of *Rhizobium* and *Agrobacterium*. Large heterogeneity was observed between the strains. The majority of *Rhizobium meliloti* and *Rhizobium trifolii* were found to be similar, while relative dissimilarities were found between strains of *Rhizobium leguminosarum* and *Agrobacterium* tumefaciens. The results suggested that grouping of these strains based on electrophoretic protein patterns are in good agreement with other results obtained by the numerical analysis.

INTRODUCTION

The present classification of *Rhizobium*, the symbiotic nitrogen fixing bacteria is based on cross inoculation between the bacteria and their various legume hosts.

This type of classification is often unsatisfactory and causes several arguments (1, 2). For example, many legume hosts are not included in the six cross inoculation groups which deine the recognized. Cross inoculation groups are not mutually exclusive and isolation of bacteria which modulate plants in more than one group is a common event (3).

It was proven that infectivity and effectiveness in *Rhizobium* were carried by plasmids (4, 5). Cross inoculation groupings would then be untenable as a basis for the classification of root nodule bacteria.

Several workers have tried other of classification based on genetic studies (6) biochemical properties (7), antibiotic sensitivity (8), cultural and morphological characteristics (9) to replace the symbiotic properties.

A number of molecular biological methods have been used for bacterial classification such as DNA base composition (10), DNA homology (11, 12)

The application of these methods have shown a powerful tool for the study of biological classification. The electrophoretic protein patterns have been success -fully applied for the identification and classification of different bacterial species such as *Streptomyces* (15) *Mycobacterium* (16) *Corynbacterium* (17) *Staphyloc-occus* (18), *Agrobacterium* (14) and *Pseudomonas aeruginosa* (19).

This work been conducted in an attempt to evaluate the importance of electrophoretic protein patterns for identification and grouping of *Rhizobium* and *Agrob -acterium* strains.

MATERIALS AND METHODS

Bacterial Strains : The strains of *Rhizobium* and *Agrobacterium* (1). in this study; and their sources are listed in table (1).

Cultural conditions : All strains were grown for 50 h at 30 °C in Erylenm -eyer flasks on Ty liquid medium (6). Fructose was added as a carbon source to a final concentration of 1%.

Cells were centrifuged at 5000rpm and suspended in 10mM phosphate buffer pH7. Cells were harvested by centrifugation and washed once in the same buffer and twice in 3mM Tris-HCl pH7.

Soluble protein extraction : 5ml of 6.4mM Tris-HCl buffer pH 8.4 containing 0.001 % Deoxyribonuc -lease (Fluka) were added to 5g/(w/w) of bacteria.

The bacterial suspension was subjected to sanction for six minutes. (2 min interval using Soniprep 150 MSE). Bacterial debris were removed by centrifugation (4°C, 30 min) at 10000rpm in a backman model J-21 b centrifuge. The supernatant was centrifuged (4°C, 1h) at 25000rpm. The supernatant was again centrifuged (4°C, 4h) at 28000rpm. The protein concentration of the clear supernatant was determined using Bradford method (20) and bovine serum albumin (faction V sigma) as standard.

Protein samples were dialyzed with one change for 48 hours against distilled water, lyophilized and resolved in 38mM Tris-glycine buffer pH8.3 and kept at -20°C until use.

Discontinuous disc-gel electrophoresis was carried out according to Khanaka (19) using 7% (w/v) acrylamide and 0.184 (w/v) N,N methylene bis-acrylamide in lower gel and 2.5% (w/v) acrylamide, 0.625 (w/v) N,N-methylene bis acrylamide in stacking gel. The gels were poured in glass tubes (5.0 x 125mm) to a height of approximately 110mm. A sample containing up to 150µg of protein was applied to each tube gel. Bovine serum albumin fraction V (80µg protein) was supplemented to one gel as reference protein in each experiment. Electrophoresis was carried out in Bio-Rad model 175 tube using 38 mM Tris-glycine buffer pH.3 in the lower and upper chambers. The temperature of electrode buffer was maintained at 8°C using Bio-Rad chiller model E4800. A constant current of 1.0mA/ gel was passed for 15 minutes with the electrode in the lower buffer connected to the anode, followed by 4.5mA/gel until the bromophenol blue band marker had reached the bottom of the tube gel.

Electrophoretic analysis of soluble protein for each strain was run at least twice in duplicate. The gels were stained with 0.1% Coomassie brilliant blue G-250 (Sigma) in Methanol/acetic acid / water (5.5 : 1 : 3.2 by vol.). The gels were sanded with photometer integrator registrator (Vernon) at wave length of 588nm.

RESULTS AND DISCUSSION

On the basic of the electrophoretic mobility, on polyacr -ylamide gel, proteins patterns from fifteen strains of *R. leguminosarum*, *R.melilotic*, *R. trifolii* and *Agrobac -terium tumefaciens* were for numerical analysis (Fig. 1).

The obtained protein patterns showed some reference bands which were

comparable on the different densitometer tracing in single runs. These peaks were numbered and examined for their presence or absence on the tracings (Fig. 2).

The reproducibility of protein bands from any strain was generally good. Fig.1 reflected an overall dissimilarity between these strains. Considerable variation in protein band patterns were observed. Some reference bands were noticed for each group. Strong similarity in protein patterns between strains L12 and L53 were observed; however a supplementary protein band was found with the strain L12. Fig.1.

More variation was found between RL17 and RL18 which indicates that strains of *R. legumin -osarum* are very heterogeneous. Similar results were observed by Legrand (22).

Three strains of *R. meliloti* 2011, AR16, M44 showed strong similarity; while more variation were found between M55 and M444; however the lack of protein observed on the two strains on the gel as well as on the densitometer tracing (Fig. 1 and Fig. 2) may be due to the presence of polysaccharide in protein solution. The production of the polysaccharide is a characteristic feature in *Rhizobium*. On the other hand previous works have shown that strains of *R. meliloti* are homogeneous (1) little similarity was found between *Agrobacterium* strains; however the genus *Agrobacterium* is known to be relatively homogeneous and showed close relationship with *R. meliloti* (9). This is likely due to the inter and infra grouping of the genus obtained through DNA-DNA hybridization (21). De-ley has also shown differences in electrophoretic protein patterns between *Agrobacterium* strains (14).

High similarity was found between two strains of *R. trifolii* T55 and T25S while both strains were clearly different from *R. trifolii* CC2480. These results were also confirmed by other results done in numerical taxonomy (22).

Protein patterns of *R. leguminosarum* L53 and *R. trifolii* T25, T55 were found to be similar. Certain similarity was found between *R. trifolii* T5 and each strain 2011, Ar16, M44; on the other hand *R. trifolii* CC240 was quite different from these three strains.

Little similarity was observed between *R. leguminosarum* L18 and *R. trifolii* T5. Legrand (22) has found identical results by the study of numerical taxonomy concerning these results.

Previous work (22, 23) has shown that *R. leguminosarum* L18 very close taxonomically to *A. tumefaciens* B6 and both are similar to R. meliloti 2011.

From this study it can be concluded that our results regarding the grouping of these strains are in good agreement with the results, previously obtained by the numerical analysis.

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of DNA-binding Ha-type proteins from Rhizobiaceae. Eur. J. Biochem. 147 : 343-349 (1985).

Table 1 : *Rhizobium* and *Agrobacterium* tested

Organism	Source*
Rhizobium meliloti 2011	RCP
Rhizobium meliloti 44	CIAT
Rhizobium meliloti 444	CIAT
Rhizobium meliloti Ar16	SRMS
Rhizobium meliloti M5S	USTL
Rhizobium leguminosarum L12	USTL
Rhizobium leguminosarum L17	FSAG
Rhizobium leguminosarum L18	FSAG
Rhizobium leguminosarum L53	RIO
Rhizobium trifolii T5	PCR
Rhizobium trifolii 25	USTIL
Rhizobium trifolii CC2480	CSIRO
Agrobacterium tumefaciens B6	RUG
Agrobacterium tumefaciens O362	RUG
Agrobacterium tumefaciens B37	RUG

*RCR: Rothamsted Collection of Rhizobium Harpenden Hertefor - dshire, United Kingdom.

CIAT: Center International Agricu - ltural Tropical de Colombia.

SRMS: Station de Recherches de Micro - biologie de Sol de Dijon, France.

USTL: Univeristy des Sciences et Techn - iques de Lille, France.

FSAG: Faculte des Sciences Agronomiques de Gembloux, Belgique.

RIO: Research Institute of Ontario, Canada.

CSIRO: Commonwealth Scientific and Industrial Research Organization of Canberra, Australia.

RUG: Rijksuniversiteit, Gent, Belgium.

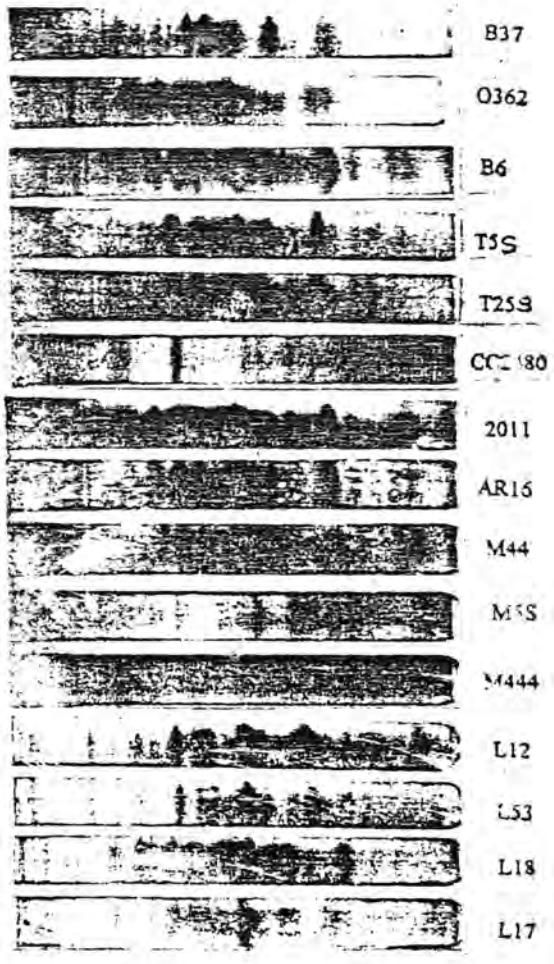

Figure 1: Electrophorograms of soluble proteins from fifteen strains of *Rhizobium* and *Agrobacterium*.

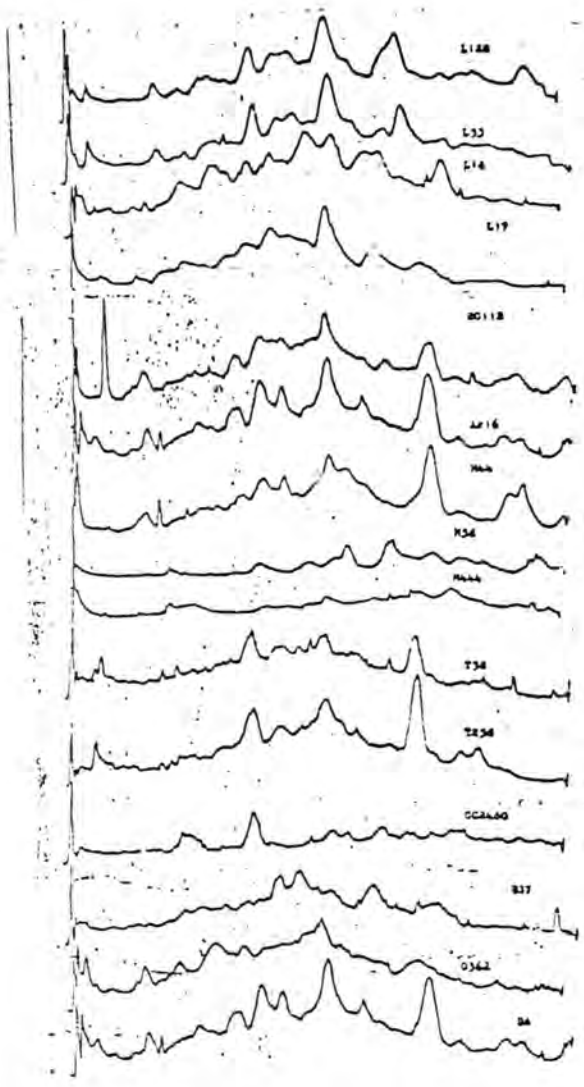

Figure 2: Densitometer tracing of the protein patterns of *Rhisobium* and *Agrobacterium* strains

Hermaphroditism in The Minnow *Phoxinus phoxinus* L.

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الخلاصة

سجل لاول مرة وجود حالة جنسية خنثية في سمكة المنوه *pHOXINE PHOXINUS* في عام ١٩٨٨. وقد تمت مناقشة اهمية هذه الظاهرة نسبة الى السلوك التكاثري والتواجد والبقاء لهذا النوع الصغير من اسماك المياه العذبة كذلك توقشت هذه الحالة مقارنة بحالات مماثلة في انواع اخرى من الاسماك الخنثية .

ABSTRACT

Hemaphroditism type of sexuality was recorded for the first time in the minnow *Phoxinus phoxinus* L. 1988 . The importance of this phenomenon in relation to the production behaviour and survival of this small freshwater species are also discussed , in comparison to the simliar observations in other Hermaphrodite species.

INTRODUCTION

The minnow *Phoxinus phoxinus* L. is small freshwater fish belongs to the family Cyprinidae species of this family are found in wide range over the world (1) . Most of the commercial Iraqi fish species belong to this family , such as *Barbus barbus* , *B. esocinus* , *B. grypus* , *B. luteus* , *B. sharpeyi* , *B. xanthopterus* , *Cyprinus carpip* and *Aspius vorax* (2) . It is well known that the species of this family are normal dioecious animals (2) . However , Hermaphroditism has been reported in some species , such as , the oviparous cyprinodont *Rivulus Marmoratus* which is capable of internal self-fertilization (3,4) . Gynogenesis has also been reported in the natural population of crucian carp (silver gold fish)*Carrassius auratus gibelio* (5). In the minnow *Phoxinus leavis* sex reversal was reported (6). Previous studies on the reproduction of *Phoxinus phoxinus* showed it as a normal dioecious fish (7,8). This study was carried out to inestigate the modification in the gonads of *P. phoxinus* during the spawning season and the posibility of hermaphroditism in this species.

MATERIALS AND METHODS

The minnows *Phoxinus phoxinus* were collected monthly from streams and small lakes in area round Keil(W.G.) , by seining with small net of 3mm. mesh , during May - July 1988 . The samples with open cavity were preserved immediatly in buffered 8% formaldehyde freshwater solution . Total length , weight and sex ratio were determined . Gonads were checked morphologically and histologically for any sign of hermaphroditism .

RESULTS

Sex ratio :

Table (1) shows the domination of females over males duringthe period of May - July . This domination increases somewhat proportionally with length.

May - July . This domination increases somewhat proportionally with length.

Hermaphroditisim :

A case of hermaphroditic gonad type was discovered for the first time in this species *P. phoxinus* . It was of 62mm. length and 2.804gm. total weight (with a gonad of 0.25ogm.). And three years age . This hermophroditic gonad consists of both testicular and suppressed ovarian parts . The oocytes are in the yolk vesicle stage of development, with the apearence of early vitellogenesis stage of yolk granules (Figs 1,2) , according to Wallaces calssification of oocyte growth stages ,(9) , which the secondary oocyte growth stages , in scot calssification of oocyte growth stages (10) . The testicular part of the hermophroditic gonad shows systs of spermatogonia, (figs 1,2).

DISCUSSION

In present study , sex ratio in *P. phoxinus* a domination of females over males . This domination is slightly higher in the larger sizes than in smaller ones . In Windermer , the differences in length between males and females of *P. phoxinus* , in their first and second years insgnificant . However the three -years old males are much smaller than the females (11) . Similar observations were reported in *P. leavis* , where no significant differences in length of males and females was noticed in their first , second and third years , while in the following year calsses , the males are appreciably smaller than females (12) . These results could explain the slight increase in the female sex ratio over males in the larger sizes . The present hermaphrodite case fell within the age of three years old . Moreover , both ovarin and testicular areas have reached appreciable stages in their development toward the maturation of oocytes and sperms , but they have not reached the final stage of maturation . However , the ovarian region acheived an advanced stage in comparison to the testicular region of the present hermaphrodite gonad . On the other hand the testicular area of the present gonad is much larger than the overian area. From the evidance in the present study , it can be concluded that it is a synchronous hermaphrodite type , in which male and female sex cells ripen at the same time regardless whether or not self-fertilization is possible , rather than consective type of hermaphroditism . In the minnow *P. leavis* intersexual gonads were found , where the overian portion was suppressed , while the testicular region was normal (6). The latter auther suggested that these intersexes represented tranitory stageschanging from female to male . In the present study , the advanced ripen of the overian region make it difficult to reach such a conclusion despite its suppressed portion , and the synchronous type of hermaphroditisim is the most convieniant type in this case . However synchronous hrmaphroditism type have been noticed in many species such as , *Serranus subligerius* , a florida sernid where self-fertilization and development are possible (13,14) . Four species of serranid fish from Bermuda , one belonging to the genus *Hypoplectrus* and three of the genus *Prinodus* are synchronous hermaphrodites , while at least nine species are protogynous hermaphrodites (15). The oviparous cyprinodont , *Rivulus marmoratus* are mostly genuine hermaphrodites capable of internal self-fertilization (3,4). According to the latter auther low temperature (18-20 °C) tends to transform the hermaphrodites to males (16). However , in the Baltic *Sprattus sprattus* sex reversal from male to female phase was noticed in the large size females (17). It seems that the responses of populations of hermaphroditic species to the size of specific mortality factors , are nicely synchronized with the gonadal

differentiation to ensure the approperiate representations in the next generations. Moreover , the minnow *P. phoxinus* is a short life-span species (11). Observations on the reproduction behaiviour of this species show many dangrous threatining their chance of survival , for both the spawners and the discharged eggs . Because females and males have to swim over gravel in the outer edge of the shallow waters and shed their eggs and milt between the stones there ,(8). From these results it can be concluded that synchronised hermaphroditism in *P. phoxinus* seems to be one of the means by which this small freshwater species compensate their heavy loss during the spawning activities , to increase their chance of survival in the reproduction cycle.

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Table (1) : Length composition and sex ratio of P.phoxinus during the spawning season (May-July).

Length mm.	Total	No. Female	No. Male
30-40	188	109 (58%)	79 (42%)
41-50	209	125 (60%)	84 (40%)
51-60	213	147 (69%)	66 (31%)
61-70	191	139 (73%)	52 (27%)
71-80	71	50 (70%)	21 (30%)
Total	872	570 (65%)	302 (35%)

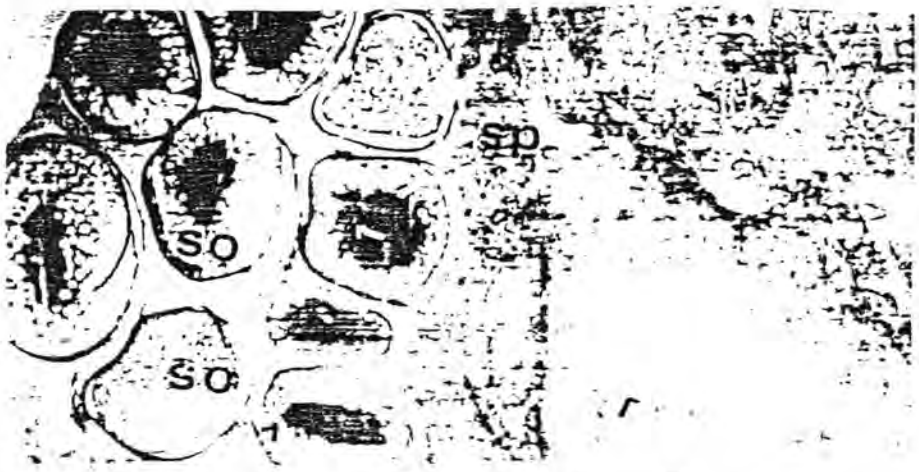

Figure (1) : The hermaphrodite gonad consist of secondary oocytes (so) in the overian part and spermatogonia in the cyst in the testicular part (sp) in *Phoxinus phoxinus* 25X

Figure (2) : The hermaphrodite gonad consist of early vitellogenesis (vo) in the overian part and spermatogonia cyst in the testicular part (sp) in *Phoxinus phoxinus* 25X

The Unscheduled DNA Synthesis (Repair) of Hyvrid Mo-1 Obtained from Somatic Fusion Between Rodent ($A_{2\,3}$) and *Xeroderma pigmentosum* Cells

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الخلاصة

درست امكانية التصليح الاستثنائي في خلايا كل من الابوين (A_{23} و XP) والهجين (Mo-1) . وقد تبين ان للهجين امكانية متوسطة في هذا التصليح . من ناحية اخرى فان قابلية الهجين باصلاح ضرر الاشعة فوق البنفسجية قد درس باستخدام تقنية التدرج السكري القلوي التي اوضحت ان للهجين قدرة عالية على التصليح مع ميل لتقطع جزيئة الـDNA وتكوين قطع صغيرة ذات وزن جزئي عالي ، في الجرع الكبيرة (٢٠٠-٤٠٠ ارك/ملم^{٢})ز دعت هذه النتائج الى افتراض انتقال تركيب جزيئي معقد لـ DNA الهجين وان علاقة هذا التركيب بالتصليح الخلوي والتسرطن الاشعاعي قد نوقش.

ABSTRACT

The unscheduled DNA synthesis (UDS) of parents (A_{23} rodent fibroblast and *Xeroderma pigmantosum* skin cells) as well as their hybrid (Mo-1) has been studied. Data revealed that the hybrid is moderate in its capacity to post replication repair. The ability of the hybvrid to repair UV-induced damage was determined using the alkaline sucrose gradient technique. The hybrid reflected an appreciable potentios of repair and tendency to forom small fragments of high molecular weight at higher UV doded. The possibility of transfer of a complex structure is inferred and the connection of these results to cellular repair and radiation carcinogenesis is discussed.

INTRODUCTION

The relationship between carcinogenic effects of ultra violet (UV) radiation and specific genetic damage is at present only tentative (1). The formation of cyyclobutyl dimmers between adjacent thymine molecules has shaown to be the major UV induced DNA damage in mammalian cells(2) and bacteria(3). These dimmers may be repaired by several enzymatic systems (4). Hence, deficient in ezcision repair system, e.g. cells from skin disease *Xeroderma pigmantosum* (XP), are extremely sensitive to UV radiation (5). In addition, these cells exhibited a reduced level of undcheduled DNA (repair) synthesis(6) and can easily transformed to neoplasia. It was therefore inferred the XP transfomation is mainly due to DNA installed photochemical lesions that cannot be repaired(7). Rodent cells, on the other hand, seem to excise very few pyrimidine dimmers compared to cultured human cells(8) and there is an evidence that they can recover UV damage via bipassing photochemical ledions during DNA replication(9). At present, one of the

powerful techniques used to study the nature of malignancy(10), tumour immunity (11), and viral transformation in culture has been the hybridization of somatic cells (12).

Presently, hoever, Al-Shaickly and Boyle (13) isolated hybrid cells (Mo-1 & Mo-2) from fusion between rodent fibroblast A_{23} and a non chromosomal repair system was interred (14). The understanding of the UV response of this hybrid on the basis of amount of damage (thyminedimmers) and capacity of repair (UDS) may facilitate the understanding of this very subtle relationship.

MATERIAL AND METHODS

1. Cell Lines : A_{23}, a fibroblast cell line derived from chinese gamster DON. It is a thymidint kinase deficient cell acquire a potent UV repair capacity. *Xeroderma pigmentosum* (XP), a human skin cell deficient of excision repair capacity. Hybrid (Mo-1 & Mo-2) cells were obtained from fusion between A_{23} and XP cells (13). Parent and hybrid cells were prepared as monolayers in 13-DMEM and HAT-Oia-5media respectively (13).
2. Sucrose Gradient Analysis of Newly Synthesised DNA:
3. UV treatment of cells: logarithmic phase cells grown in 13 DMEM were labelled with ^{14}C thymidint (0.2 μCi/ml of 57 mg/mM specific activity). The cells (approx. $5x10^5$ cell/ml) were washed twice by centrifugation and resuspension in fresh medium. 5ml aliquotes, plated in 60 mm Falcon's Petri-dich, were allowed to attach for 4h at 37°C. Attached cells were then washed with sterile PBS`` and UV irradiated dry, in plates with lids open (14). After irradiation cells were trypsonized, neutralised by 13-DMEM and centrifuged to prepare a concentrated suspension in physiological saline. Control cells (i.e. those not treated with UV) were further incubated for 30 min while treated cells were incubated for 60 min.
4. 2-Preparation of gradients: 14.4ml isokinetic gradients of 5-34.77% sucrose were generated in 16.6ml polypropyleneMSE centrifuge tubes using an Isograd Gradient Maker (Baird & Tatlock). The sucrose (Analar grade TNA as free) was dissolved in 0.01 tris HCl, 0.001M EDTA, 0.1M NaCl, 0.01M tris HCl, pH 12.5.
5. Centrifugation: cells treated with lysing solution (0.2 ml of 0.05 M NaOH, 0.95 M Nacl and 0.01 M EDTA) were centrifuged at 25000 rpm (100,000 G basedon radius maximum) at 21 °C using 6 x 16.5 ml aluminium swing out rotor in MSE 65 Ultracentrifuge. After centrifugation the gradients were fractionated into 30 x 11 drop fraction. To each fraction, 5 ml ice cold 10% trichloro acetic acid was added followed by 100 μg of crude DNA as a carrier. The fractions were allowed to settel at 4 °C, overnight, before cold TCA precipitates were collected. Precipitates were washed with 2.5% cold TCA, dried under infra red lamp and the radioactivity measured in 4 ml toluene based phosphor containing PPO (4 g/1) and POPOP (0.1 g/1) in the liquid Scintillation Counter (Perkin Elmer Beta S.Counter).
6. Determination of molecular weight: $S_{20}IW$ were calculated according to equation
7. $S_{20}IW = K. D/w^2t$ Burgt and Hershey (16)
8. where K is the calibrated constant determind by measuring the distance D (meters) segmented in t seconds during centrifugation at w radius per second. The average molecular weight (Mi) of DNA molecules in each fraction was then determined using the relationshop:
9. $S_{20,w} = 0.0528\ Mi^{0.4}$

10. The molecular weight of each fraction was used to calculate the number average molecular weight (Mw) for the region of each gradient which contained the radioactive peak. Calculations were performed by computer and were based on the method described by Ehmann and lett (17).

Assay of Pyrimidine Dimmers:

Amounts of thymidine dimmers produced in DNA following UV exposures was determined using (18) method. It involved the labelling of DNA with ^{3}H-Thymine and measuring ratio of dimmers to the total DNA thymine using double dimensions paper chromatography technique. Solvents adopted in this technique were:

N butanol and water (1/5 v:v) for the first dimension

Ammonium sulphate, sodium acetateisopropanol (1/10 v:v) the seconed dimension. This method facilitate the dimmer detection at the level of 0.02%.

RESULTS AND DISCUSSION

Assay of Thymine Dimmers in UV Irradiated:

Amounts of thymine dimmers produced at time of irradiation (T_0) and 24th after irradiation (T_{24}) for parents (A_{23} & XP) and hybrid (Mo-1) cells are depicted in Fig. 1. The percentage dimmers produced at T_0 are the same for parents as for hybrid cells. However, amount of repair of UV damage 24th after irradiation (T_{24}) vary among parents as well as hybrid cells. The least repair capacity (represented by percentage removal of dimmers) is seen with XP (less than 2%), and most with A_{23} (approx. 46%). Mo-1 however, showed about same repair capacity as A_{23} (approx. 40%). These results showed that the initial absorbed dose of UV produce same amounts of dimmers in cells karted. Dimmers decreased to various levels (except that of XP) 24th after irradiation indicating variable efficiencies among cells tested. This finding is in consistence with Setlow and Carrier (19). The hybrid capacity for UDS is moderate in comparison to both parents which point out the possibility of transfer of genetic marker associated to excision repair (post replication repair) to hybrid from A_{23}. It will be of interest to further investigate this fact, most likely, by marker rescue and transfer test; when one or both parents irradiated before cell fusion.

Sedimentation Profiles of UV Irradiated Mo-1 Cells:

The isokinetic sedimentation profiles of Mo-1 cells centrifuged following UV irradiation (25, 50, 100, 200, 300, and 400 ergs/mm^2) at T_0 , T_2, and T_{24} hours after irradiation are depicted in Fig. 2, 3, 4, 5, and 6. The unirradiated cells exhibit a broad unidal sedimentation profiles with average molecular weight corresponding to 8.99 x 10^8 daltons and peak sedimentation at fraction number ranging between 14 to 18. The broadness of profile represent (presumably) the labelling of DNA replicons derived from the early stages of replication to those which are almost completed. At T_0, the same sedimentation profiles were noticed at all UV doses given, but, nuclear materials showed an apparent tendency to from shorter fragments at doses above 300 ergs/mm^2. Two hours post replication incubation (T_2) revealed, once again, this tendency and fragments of low molecular weight were notably detected (at doses 200 and 400 ergs/mm^2) with peak sedimentation at fraction 10 (towards the bottom of the gradient indicating an increment of the size of nasant DNA pieces synthesised after irradiation or due to fractions of a complex associated with nucleon prokeines or lipids. After 24 hours

incubation (T_{24}) a pronounced peak shift is seen when cells irradiated at low UV doses (Fig.3 & 4). It is also clear that high molecular weight and shorter fragments still sedimenting towards the bottom of the gradient at doses above 200 ergs/mm^2; and Nuclear materials seemed to segregate to small fragments.

Analysis of Mo-1 isokinetic sedimentation profiles indicate the ability of this hybrid to repair UV induced thymine dimmers. It also, revealed the apparent tendency to be fragmented at fast, then sedimented rapidly towards the bottom of the gradient. This may be due to high molecular weight fragments that arises from combination with nuclear proteins. A similar suggestion was made by Fox et al. (20), and Fox (21).

In placing the results pf present study in prospective to radiation carcinogenesis, cell fusion between XP and A_{23} cells has permitted the repair of UV damage relevant to theoretical proposition made in the introduction. The transfer of genetic marker(s) associated with repair during fusion could alternatively have some bearing on oncology and possible future treatment of the human skin cancer *"Xeroderma pigmentosum"*.

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FIGURE LEGENDS

1. Figure 1: Percentage dimmers produced in parent (A_{23} and XP) and hybrid Mo-1 cells after exposure to UV radiation at T_0 and T_{24}h post irradiation incubation.
2. Figure 2: Alkaline sucrose gradient sedimentation profiles of Mo-1 cells UV-irradiated with 25,50, and 100 ergs/mm^2 and assayed immediately after irradiation.
3. Figure 3: Alkaline sucrose gradient sedimentation profiles of Mo-1 cells UV-irradiated with 200, 300, and 400 ergs/mm^2 and assayed immediately after irradiation.
4. Figure 4: Alkaline sucrose gradient sedimentation profiles of Mo-1 cells UV-irradiated with 50 and 100 ergs/mm^2 and assayed after 2h post-irradiation incubation.
5. Figure 5: Alkaline sucrose gradient sedimentation profiles of Mo-1 cells Uv-irradiated with 200 and 400 ergs/mm^2 and assayed 2h after irradiation.
6. Figure 6: Alkaline sucrose gradient sedimentation profiles of Mo-1 cells UV-irradiated with 25, 50, and 100 ergs/mm^2 and incubated 24th before assay.
7. Figure 7: Alkaline sucrose gradient sedimentation profiles of MO-1 cells UV-irradiated with 200, 300, and 400 ergs/mm^2 and assayed 24th after irradiation.

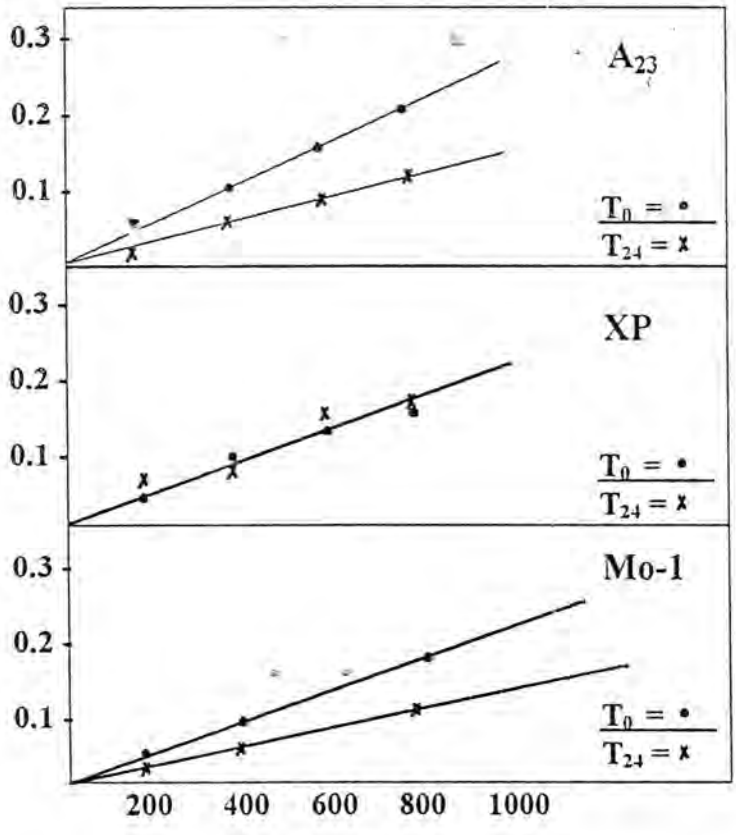


Fig. 1

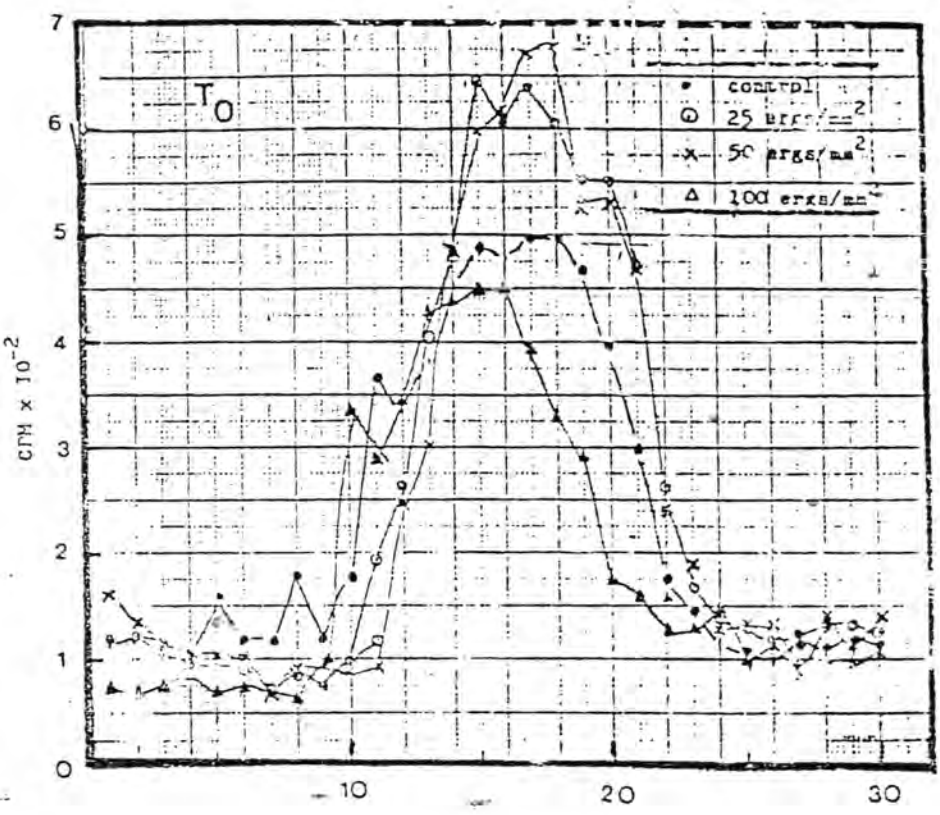
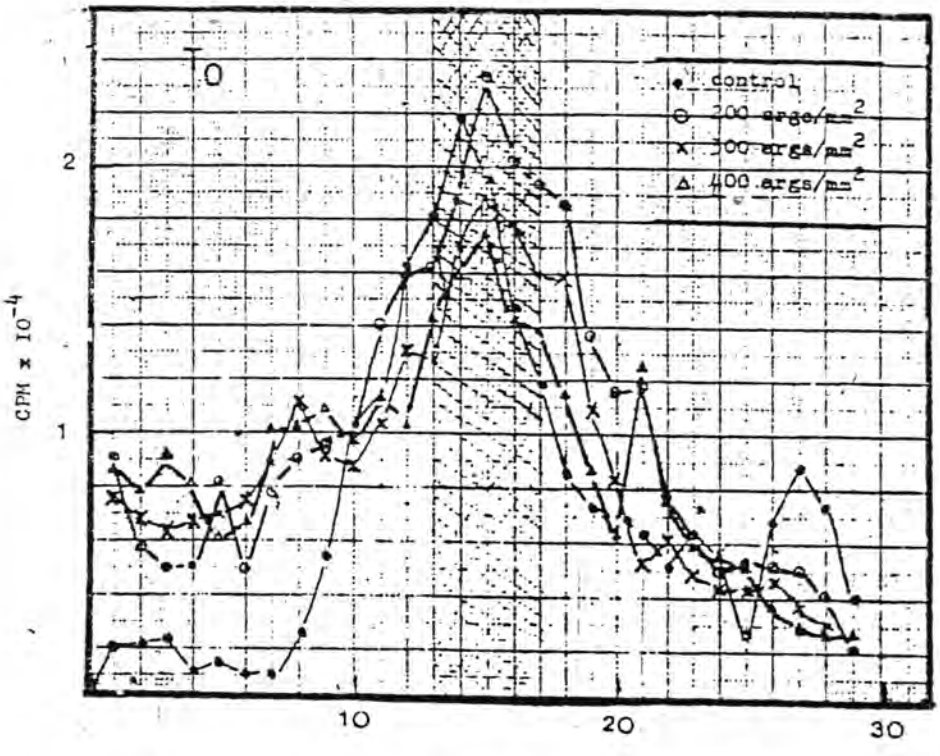


Figure 2 Fraction Number



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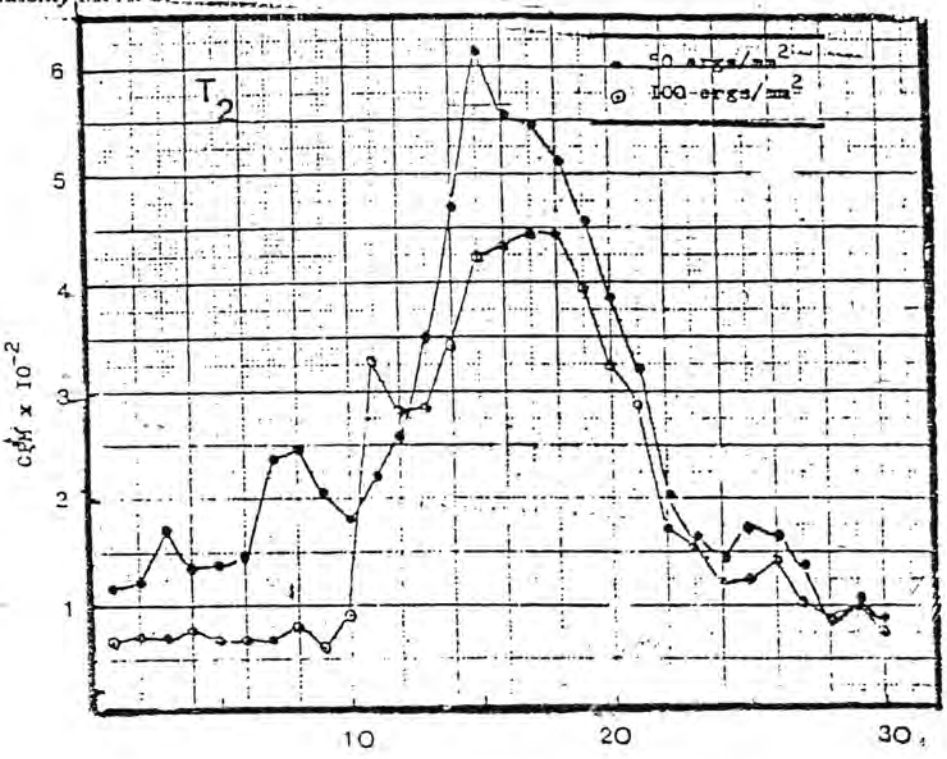

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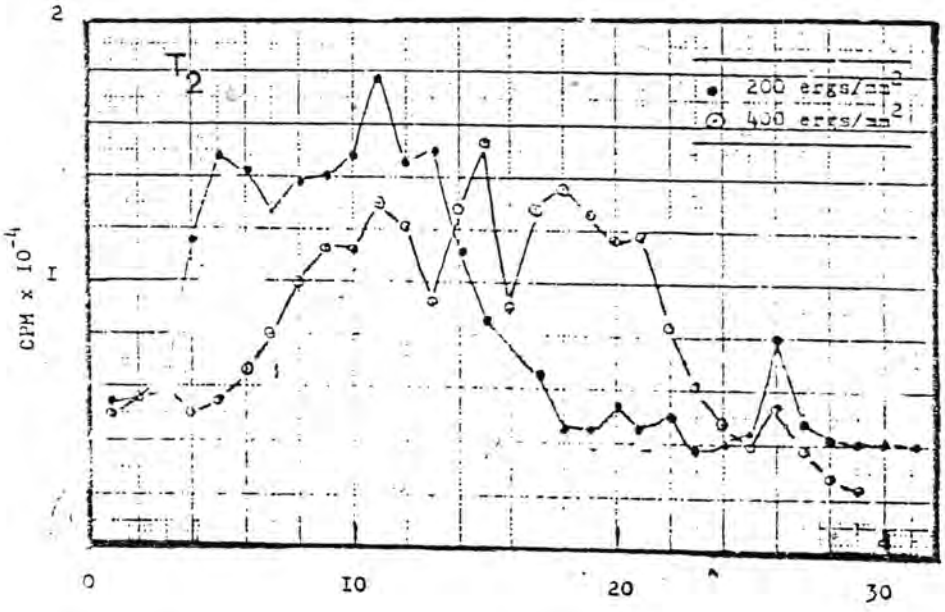

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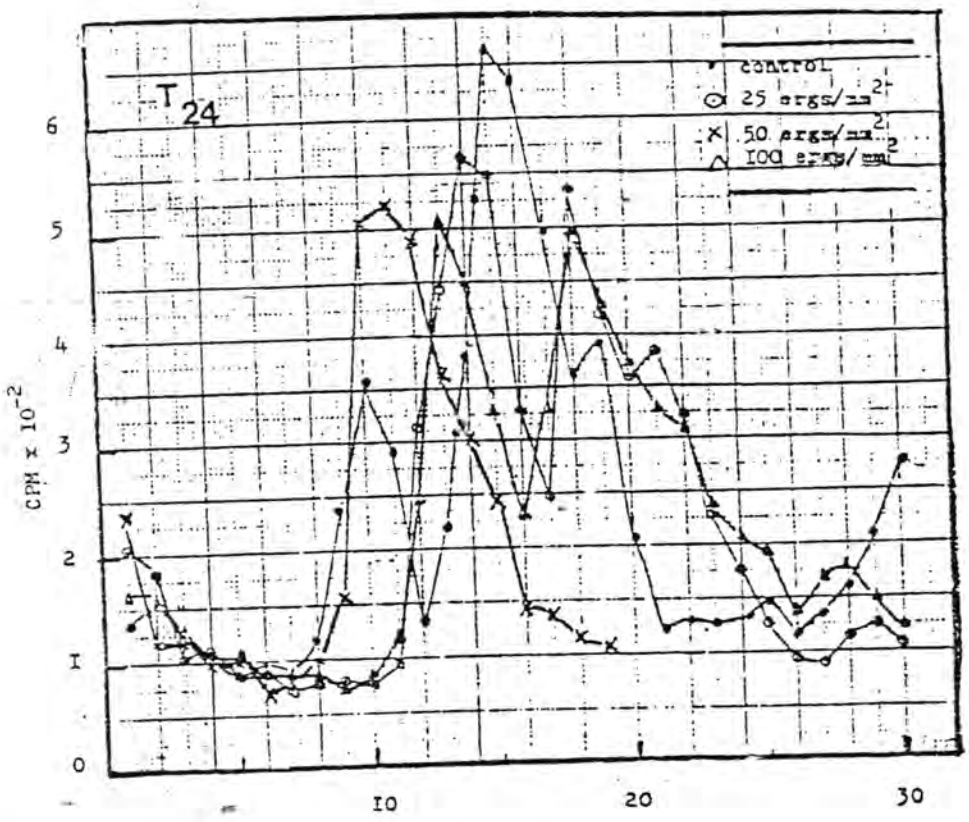

Figure 6 Fraction Number

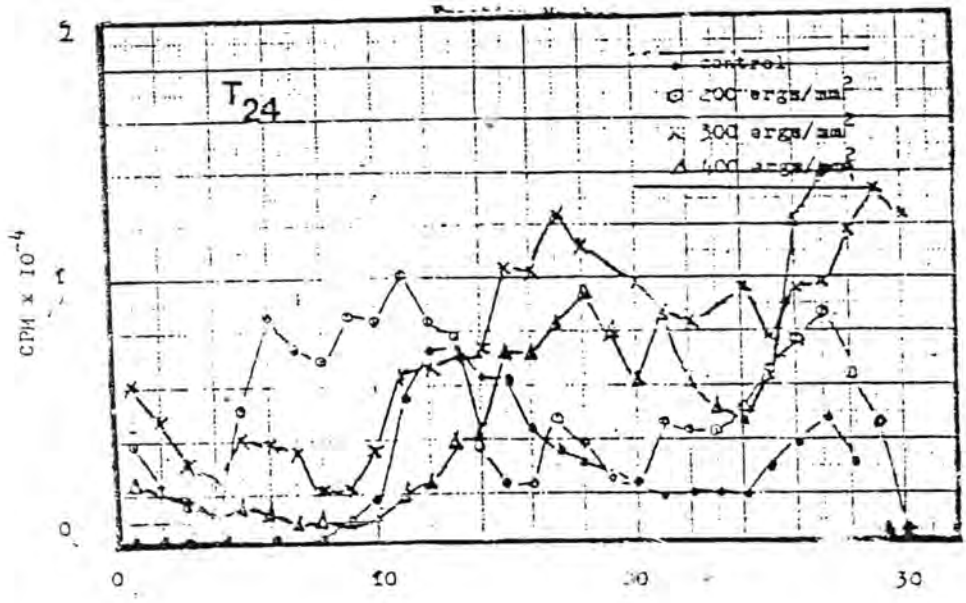

Figure 7 Fraction Number

Transposon Mutagenesis in Rhizobium Meliloti Affecting on The Production of Exopolysoccharides

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الخلاصة

باستعمال البلازميد الانتحاري كناقل فقد ادخل الترانسبوزون Tn5 الى جينيوم R. meliloti سلالة M28 المقاومة للستربتومايسين. وجد ان Tn5 يرتبط مع الجينات المسؤولة عن تمثيل السكريات المتعددة . فقد عزل من الذرية العشوائية التي تحمل Tn5 0.3% من الطفرات ناقصة التغذية . ان انتاج السكريات المتعددة لكل من المرتدات الكاذبة كان ضعيفا وذلك عند زرعها على وسط الفركتوز IS بينما على وسط الفركتوز RC انتجت المرتدات الكاذبة (3.22) مقدار 50% من السكريات المتعددة بالمقارنة مع السلالات البرية . واشارت النتائج بان التعبير الضعيف للجينات المشفرة لانتاج السكريات المتعددة لم تؤثر على قدرات الطفرات التكافلية .

ABSTRACT

Using a suicide plasmide as a vector, the transposen Tn5 was introduced into genome of R. meliloti M28 Str^r. Tn5 was found to integrate into the genes involved in the synthesis of polysacchoride. Among clones carrying random Tn5 insertion, 0.3% auxotrophes mutants was isolated. The production of exopoly saccharides produced by the wild type strain. Results indicated that the weak experession of the genes coding for the synthesis of exopolysaccharides did not disturb the symbiotics properties of mutants.

INTRODUCTION

Transposons are discrete sequences of DNA that are incapable of silf - replication and can insert into many sites of bacterial genomes, often giving rise to the formation of strongly polar mutation[1]. The transposon Tn5 conferring Kanamycin - Neomycin resistance [2,3] has been used as a biological mutagen to select auxotrophic mutants of *Escherichia coli*, a defective prophage λ serving as a vehicle of Tn5 [1,4].

In some non - enteric gram negative bacteria, however, introduction of Tn5 has been obtained using a suicide plasmid harbouring both the phage Mu DNA and various transposons. The presence of Mu DNA has been shown to prevent the maintenance of the vector in the recipient bacteria. This is probably due to an increase of host restriction on the hybrid plasmid and to the expression of at least one undetermined function of Mu[5,6,7]. Therefore, the phenonmenon of such plasmids transferring but not establishing allows the translication of transposons from these plasmids to the integration sites in the genome of their new hosts.

Recently this mutagenesis procedure has been employed to generate Tn5 - induced auxotrophic[7,8,9,10], carbohydrate [11], succinate transport[12] and sumbiotically defective[9,13,14]. The specificty of Tn5 insertion to the *E. coli* genome has been reported[1,4]. The destribution of insetion sites is not perfectly random, because certain genes

perfectly random, because certain genes appear to be hot spots at which insertion events occur repeatedly and exccessively. In some strains of fast growing Rhizobium species, Tn5 inserts at many sites, showing a low specificity of integration in their genome[7].

The aim of this work was to study the effect of Tn5 mutagensis on the production of exopolysaccharides in relation to symbiotic properties in *Rhisobium meliloti*.

MATERIALS AND METHODS

Bacterial strains

Escherichia coli strain SIII harbouring the ruicide plasmid PSP601 was used as a donor of Tn5, its phenotype is thi`, thr`, leu`, lac`, sup^F, his: Mu_{ets} Tn_1 (Amp), Tn5 (Km) Tn402 (Tp) Tn1771 (Tet).

Rhizobium meliloti, strain M28 str. (spontaneous sm^r. mutant) was obtained from the laboratory of microbiology univ. of Lille, France. This strain was fully effective in symbiosis with alfalfa *Medicago sativa*.

Media

Liquid cultures of *E. cole* and *R. meliloti* strains were grown at 30 °C in Luria broth (15) and trypatase yeast extract TY medium (7) respectively. Rhizobium complex solid medium (Rc agar) and Rhizobium minimal medium were prepared as in (16).

Transposon mutagenesis

The culture of the donor *E. coli* SIII PSP601 and recipient *R. meliloti* strain M28 str. were grown to late exponential phase and mixed at a ratio 1:1. The mixture was filterde through a sartorius membrane (0.2 μm porosity) which was incubated overnight on T_y agar at 30 °C.

The bacteria were then washed, diluted and plated on to RC agar supplemented with strptomycin sulphate 400 μg/ml (sigma) and kanamycin sulphate 200 μg/ml (sigma). This medium enabled us to counterselect against the donor and to isolate Tn5 carrying *R. meliloti* derivitives. These were detected after three days incubation at 30 °C.

Characterization of the auxotrophic mutant:

For detection of auzotrophic mutaauts, separate colonies grown on RC agar supplemented with streptomycin and kanamycin were replica plated onto R agar supplemented with various amino acids 20 μg/ml. The mutants were further analysed for their ability to grow on R agar containing different amino acids.

Agarose gel electrophoresis:

Detection of plasmids in agarose gel was carried out as described by Simon [17]. Molecular weight determination was carried out according to (18).

Plants tests:

Nodulation tests were performed by inoculating bacteria onto aseptically grown alfalfa (Medicago sativa) seedlings on nitrogen free agar medium as described by Rolfe(14). Nitrogen fixation test was performed by the acetylene reduction assay as described by Dilworth (20).

RESULS AND DISCUSSION

Escherichia coli strain SII PSP601 was crossed with *R. meliloti* strain M28 str. After mating Sm^r Km^r *R. meliloti* exconjugants appeared at frequency $1.5x10^{-6}$ while the frequency of spontaneous Km^r mutation was $2.6x10^{-9}$.

Approximately 12000 Str^r Km^r exconjugants from the mating was purified on the same selective medium and relpica plated onto R. agar. About 0.3%

auxotrophs were scored among colonies carrying Tn5 insertion 28 auxotrophic mutants were obtained which were all senditive to Amp. µg/ml.

The excoujugant M 28 Str. : Tn5.3 obtained by transposition and incapble of synthesizing tryptophan was used for the isolation of protorophic mutants. The reversion frequency was 10^{-8}. Among the prototrophic mutants, two clones 3.17, 3.22 with altered colony type were obtained. Notice that these two prototrophic mutants will soon regain the mucosity after being conserved in glycerol 20% at -20 °C. The production of polysaccharides on the proliferation media measured by chemical dosage was listed in table 1. Results showed no significant differences for all different strains on TY medium, while on the Fructose RC medium the pseudorevertant 3.22 produced about 50% of the quantity of exopolysaccharides produced by the wild type strain M 28 Str. and the auxotrophic mutant M28 Str. : Tn 5.3 finally on the Fructose IS medium the production of exopolysaccharides for both strains of pseudorevertants was very weak comparing to parental strains.

In order to precise the quantity of exopolysaccharides secreted by these two pseudorevertants 3.17, 3.22 the synthesis of exoploysaccharides was subjected to non proliferation medium in the presence of labelled Fructose with C^{14}. Results have shown that the quantity of Fructose with these two pseudorevertants were 3.5 times less than the wild type strain M28 Str. and 8 times less than the auxotrophic excoujugant M28 Str. : Tn 5.3. On the other hand the strain of *E. coli* C600 has produced a negligible quantity of exopolysaccharides. Table 1 has also shown that the resistance to kanamycin was 200 µg/ml for the auxotrophic mutant M 28 Str. Tn 5.3 while the resistance to kanamycin for the two pseudoreverant 3.17, 3.22 was more than 500 ug/ml. The weak production of exopolysaccharides might be explained by the insertion of the Tn5 on the genes coding for the synthesis of exopolysaccharides. To ascertain this hypothesis, the two pseudorevertants as well as the wild type M 28 Str. and its mutant M28 Str. : Tn 5.3 for their plasmid contents. The agarose gel electrophresis fig. 1 showed that the two pseudorevertants as well as the wild type M 28 Str. and its mutant M 28 Str. Tn 5.3 carry two plasmids. The molecular weights of the two pseudorevertant were of the same order as the wild type and its mutant. The molecular weight for both plasmids were estimated 300 and 133 Md. respectively. The transconjugant M28 Str. : Tn 5.3 and the two pseudorevertants were tested for their symbioltic properties for (nodulation and nitrogen fixation) Table 2. Results indicated that the two pseudorevertants were not effected neither for infectivity property nor for the effectivness. This indicated that the weak expression of the genes coding for the synthesis of exopolysaccharides did not disturb the efficiency of these mutants. It can be concluded that transposon Tn can be grafted into the genes involved in the synthesis of exopolysaccharides.

ACKNOWLEDGEMENT

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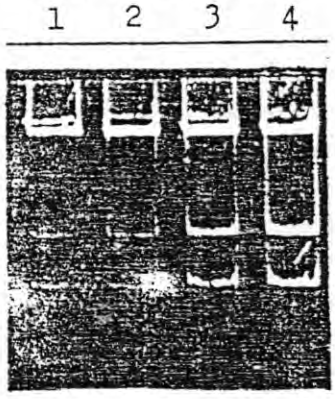

Figure 1: Plasmid content analysis of Rhizobium strains. Lane 1: M28 Str. Lane 2: M28 Str. Tn5.3, Lane 3: 3.17 Lane 4: 3.22

Table 1 : Production of exopolysaccharides by different strains of Rhizobium

Strain	Proliferation media (chemical) dosage			Medium	Phenot-ype
	TY	Fruct-ose 1% RC	Fruc-tose 1% IS	Prolife-ration Fructose 1% RN	
M28 Str.	46,9	104.1	173.5	63	Sm^r
M28 Str.:Tn 5.3	55.9	95.5	252.3	120	Sm^r Km^r 200µg/ ml) Trp
3.17	50.5	80.3	39.6	20	Sm^r Km^r µg/ ml)
3.22	60.8	44.2	37.3	16.3	Sm^r Km^r µg/ ml)
E. coli C600	ND	ND	ND	4.5	Trp^- Lea^- rk^- mk $Reca^-$ Rif^r

ND : not determined

Table 2 : Evaluation of the infectivity and effectiveness of pseudo-revertants affected on the metabolism of polysaccharides

Inocul-ation of plantule of	Dry wight of plntule /mg	Quantity of C_2H_2 trans-formed to C_2H_4 plantule nm/hr	e no. of nodule/ plantule	Isolat-ion of strain from nodule	Pheno-type
Wild strain M28 Str.	84	28,4	7	+	Smr
Auxo-trophic mutant	77	45,8	6	+	Sm^r Km^r 200µg/ m1) Trp^-
M28 Str. Tn5.3 vetant 3.17	73	18,8	11	+	Sm^r Km^r 1500µg /m1)
vertant 3.22	75	26,3	11	+	Sm^r Km^r 1500µg /m1)
Control	0	0	0	+	

Study on The Bacterial Wound Infections

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الخلاصة

من هذه الدراسة لوحظ ان بكتريا المتقلية (Proteus) المكورات العنقودية Staphylococcus, الزوائف Pseudomonas, والبكسيلا Klebsiella مثلت غالبية ٣١٠،٩٠٪ ، ١٨،١٪ ، ١٧،٤٪ ، ١٥،٧٪ على التوالي . بينما كانت بكتريا المتقلبة (Proteus) , الكلبسيلا Klebsiella والزوائف سبب خمج معظم الجروح الجراحية وكانت نسبتها ٣٦٪ ، ٢١٪ و ١٩٪ على التوالي . ان نسبة العزلات المختلطة في مستنبتات عينات الجروح الخمجية الرضية اكبر مما هي عليه في عينات الجروح الخمجية الجراحية . ان عزلات الكلبسيلا Klebsiella والزوائف Pseudomonans من كل الجروح اعطت نسبة عالية من المقاومة ضد غالبية المضادات الحياتية المستعملة .

ABSTRACT

From this it was found that *Proteus spp. Staphylococcus spp., Pseudomonas spp.* and *Klebseilla spp.* represented the main isolates of different infected traumatic wound cultures. Their percentages were 31.9%, 18.1, 17.4 and 15.7 respectively. On the other hand *proteus spp. Kebseilla* spp. and *Pseudomonas spp.* were superimposed in the surgical wound infections with 36, 21 and 19 percent respectively. The percentage of mixed isolates were higher in traumatic wound specimens than the surgical type of wounds. *Klebseilla spp.* and *pseudomonas spp.* isolated from both types of wounds showed high resistance against the majority of antibiotics were used.

INTRODUCTION

Numerous organisms may be involved in wound infections depending on type of wound itself. Weather it is surgical or traumatic wound also it depend on the cleanliness of the wound itself (1, 2, 3). Surgical wound infections are mostly nosocomialy accepted, it was found that opportunistic Gram-negative bacteria are able to multiply in the tissue of impaired defense mechanisms of the host (4). Recently it was found that the prevalence of certain Gram-negative bacteria is higher than Gram-positive bacteria in both surgical and traumatic wound infections (2, 3, 5).

Watt and Collee (6) found that it is important to do certain prophylactics to the accidental wounds with Penicillin to reduce the risk of gas gangrene and tetanus, This study was devoted to show the difference in the bacterial flora of traumatic and surgical wounds and their antibiotics resistance in Saddam General Hospital Ramadi City.

MATERIALS AND METHODS

Specimens Collection :

Different traumatic wound specimens were obtained by rubbing the

wound with sterile disposable cotton swabs moistened in sterile normal saline, (200) specimens were obtained equally from males and females.

Another (200) specimens were obtained from different postoperative infected surgical wounds from the patients admitted to the surgical wards of Saddam General Hospital in Ramadi City. Anti microbial therapy stopped 24 hours before specimens collection.

B- Isolation of Bacterial Flora :

The swabs were cultured asepticaly within one hour on the following media :

1- Blood agar (prepared from Blood base (Oxide) with 10% human blood).
2- MacConkey agar and Mantel salt agar (Oxide) incubated under aerobic conditions at 37 C° for 24 to 48 hours.
3- Blood agar plates incubated under anaerobic conditions at 37 C° for 24 to 48 hours using the anaerobic Jar and gas packs (Oxide).

Cooked meat broth (Oxide) also was employed.

C- Bacterial Identification :

The following standard tests were used for bacterial identification according to the methods of Cruskshank, et al. Finegold, et al (7, 8).

A- Morphological with Gram stain .

1- Direct smear stained with Gram stain.
2- Cultural characteristics afers 24 hours.

B- Biochemical tests .

1- Indole. 2- Urease.
1- Methy / Red, Vogus proskauer M.R. V.P.
2- Growth in KCN medium.
3- H_2O Production using kligler iron Medium.
4- Gelatinase .
5- Optochin test.
6- Oxidation or Fermentation of glucose in OF medium .
7- Nitrate reduction.
8- Citrate utilization using Simmon-citrate agar.
9- Phosphatase.
10- Sugar fermentation tests using sugar medium base with 1 % of the following sugars (Lactose, glucose, sucarose, fructose, Mannitol).
11- Oxidize test using (Fluka) Oxidize reagent.
12- Coagulase test. Slide and tube Method using sterile fresh Rabbit plasma.
13- Catalase test.

C- Other test :

1- Motility test using hanging drop method.

D- Antibiotic Susceptibility Test :

The Baure-Kirby disk diffusion method (9) was used to detect the antimicrobial susceptibility of the organisms isolated.

Biomerieux antibiotic disks were used for these tests as mentioned below.

Ampicillin	10 mgAm.
Pencillin	10 IU. P
Carbencillin	100 µg CB
Cephalexine	30 µg Cf
Chlorumphenicol	30 µgC
Gentamicin	10 µg Gm
Refampicin	30 µg Ra
Tetracycline	30 µg TE
Erthromycin	15 µg E
Lincomycine	2 µg L
Cloxacillin	5 µg

RESULTS

It was found that 68% of the traumatic wound specimens were positive, while 49% of the surgical wound specimens showed positive cultures. Results also showed that the incidence of the traumatic bacterial wound infections were as follows :

Proteus spp. 31.9%, *staphylococcus spp.* 18.1% *pseudomonans spp.* 17.4% and *klebseilla spp.* 15.7% (Table -1), while in case of surgical bacterial wound infection, the incidences were as follows ; *proteus spp.* 36%, *klebseilla spp.* 21% & *pseudomonas spp.* 19% (Table -2). Gram-negative bacteria were isolated mainly from both traumatic (76.13%) and infected surgical wounds (85.1%) (Fig. -1). No significant difference was found between males and females in the bacterial species and the number of isolates in both traumatic and surgical wounds ($p > 0.05$).

The mixed bacterial infections of traumatic wounds was higher than that of surgical wounds, their percentages were 62.5, 43.8 respectively (Table -3). *Klebsiella pneumonae* and *Pseudomonas aurogenosa* showed high resistance to the different antibiotics used (Table -4).

Table 1 : Incidence of bacterial infections in the positive pure and mixed cultures from the infected traumatic wounds.

Bacterial Isolates	No. of Pure Isolates		No. of Mixed Isolates		Total	
	Females	Males	Females	Males	No.	%
Klebsiella pneumonae	3	4	14	18	39	15.7
Pseudomonas aurogenosa	3	4	19	17	43	17.4
Proteus mirabilis	2	3	32	28	65	26.3
Proteus rettegri	1	1	7	5	14	5.6
Escherichia coli	2	1	13	10	26	10.5
Enterobacter cloacae	1	1	2	0	4	1.6
Citrobacter freundii	-	1	-	-	1	0.4
Staphylococcus aureus	4	7	11	13	35	14.1
Staph. Coagulase negative	2	3	3	2	10	4.0
Beta hamolytic Strept	2	4	0	0	6	2.4
Clostridium perfringens	1	1	1	1	4	1.6
Total	21	30	102	94	247	100

Table 2 : Incidence of bacterial infections in the positive pure and mixed cultures from the infected surgical wounds.

Bacterial Isolates	No. of Pure Isolates		No. of Mixed Isolates		Total	
	Females	Males	Females	Males	No.	%
Klebsiella pneumonae	6	5	10	10	31	21.0
Pseudomonas aurogenosa	3	4	12	9	28	19.0
Proteus mirabilis	9	10	14	10	43	29.2
Proteus rettegri	3	1	3	3	10	6.8
Escherichia coli	2	1	5	5	13	8.8
Staphylococcus aureus	2	4	2	4	12	8.8
Coagulase negative staph.	1	2	1	3	8	4.7
Beta hamolytic Strept	1	1	0	0	2	1.3
Total	27	28	48	44	147	100

Table 3 : Incidence of mixed bacterial isolates in both traumatic and infected surgical wounds.

Bacterial Isolates	Trumatic Wounds Isolates		Surgical Wounds Isolates		Total	
	Females	Males	Females	Males	Females	Males
Klebsiella spp. + Proeus spp.	7	8	5	5	25	195
E. coli + Klebsiella spp.	2	4	1	2	9	7.0
Pseudomonas spp. + Kleb. spp.	3	3	4	2	12	9.3
Klebsiella spp. + staph. spp.	2	3	0	1	6	4.6
Proteus spp. + E. coli	9	5	3	2	19	14.8
Proteus spp. + Pseudomonas spp.	8	7	6	2	23	17.9
Proters spp. + Staph. spp.	2	3	2	1	8	6.2
Pseud. spp. + Staph. spp.	3	2	1	2	8	6.2
Pseud. spp. + E. coli	1	0	0	0	1	0.7
Pseud. spp. + Proteus spp. + Staph.	4	5	1	3	13	10.1
Entero bacter + Proteus spp.	2	0	0	0	2	1.5
Clostrisium spp. + E. coli	1	1	0	0	2	1.5
Total	44	41	23	20	128	100

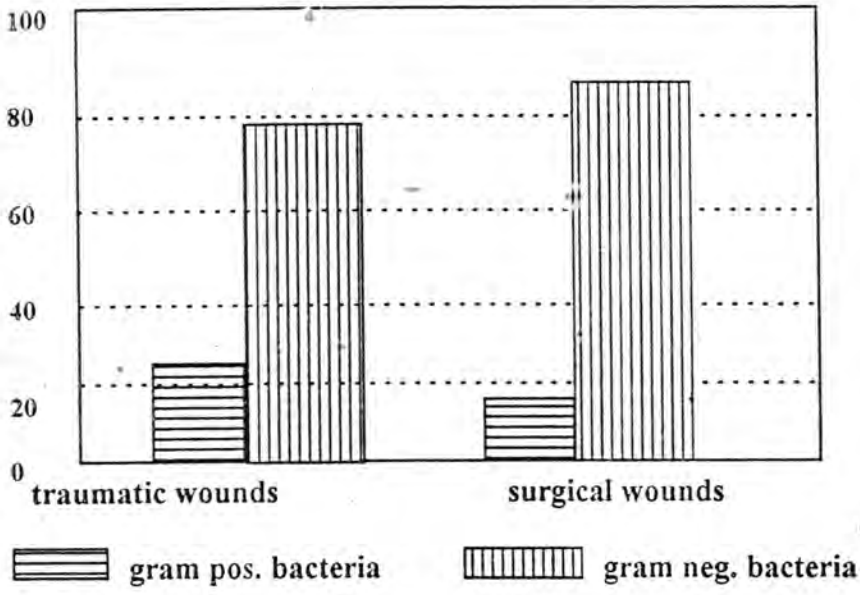

Figure 1: percentage of Gram positive & gram negative isolates

Table 4: Antibiotic susceptibility of bacterial isolates from traumatic and surgical wounds

Antibiotic	% Of Resistant Bacterial Isolates																										
	Pseudomonas			*Proteus*			*Klebsiella*			*E.Coli*			*Staphyiococi*			*Streptococci*			*Citrobacter*			*Enterobacter*			*Clostridium*		
	Tr.	Sur	Tot	Tr.	Sur	Tot	Tr.	Sur	Tot	Tr.	Sur	Tot	Tr.	Sur	Tot	Tr.	Sur	Tot	Tr.	Sur	Tot	Tr.	Sur	Tot	Tr.	Sur	Tot
Ampicillin	76	26	93	43	22	65	53	42	95	41	35	76	20	18	38	7	5	12	90	*	90	67	*	67	60	*	60
chlorum-phenicol	45	27	72	33	25	58	35	44	97	28	25	53	13	10	23	10	13	23	29	-	29	45	-	45	5	-	5
Gentami-cin	21	13	34	11	9	20	30	20	50	10	8	18	22	19	41	13	15	28	48	-	48	15	-	15	65	-	65
Cephalex-in	48	46	94	18	12	40	31	27	58	27	31	58	24	19	43	9	11	20	89	-	89	80	-	80	35	-	35
Refampic-in	27	21	48	23	18	41	8	11	19	13	16	29	10	12	22	5	3	8	27	-	27	20	-	20	15	-	15
Teracycli-ne	40	42	82	37	33	70	32	28	60	29	35	64	24	28	52	40	35	75	59	-	59	45	-	45	80	-	80
Cloxacill-in	60	30	90	47	34	81	49	41	90	43	37	80	11	9	20	11	14	25	98	-	98	90	-	90	35	-	35
Lincomy-cine	49	45	94	38	42	80	42	49	91	49	36	85	21	10	31	24	16	40	96	-	96	92	-	92	20	-	20
Erythro-mycin	45	50	95	58	34	92	40	58	98	47	45	92	23	26	49	12	8	20	95	-	95	94	-	94	15	-	15
Penicill-in	50	40	90	67	23	90	50	45	95	42	48	90	37	38	75	19	16	35	99	-	99	97	-	97	45	-	45
Carbenci-llin	23	18	41	17	18	35	47	33	80	28	22	50	24	26	50	39	48	87	50	-	50	45	-	45	60	-	60

Tr. Trumatic .
Sur. Surgical .
*** Not Isolated & not tested .**

DISCUSSION

The high percentage of positive traumatic wound specimens of both sexes was attributed to the wound sepsis with high counts of different types of bacteria during truma induction. On the other hand opposite results were seen in the surgical wounds which might be due to the lowered probability of sepsis during surgical operations and the preoperative antibiotic prophylactics (4, 5, 10). The high incidence of gram negative isolates from both traumatic and surgical wound specimens was also observed. (2. 3. 5. 11), this is probably due to the wide distribution of these bacteria and their resistance to the different environmental conditions. Traumatic and surgical wound infections were found mostly due to *Proteus spp.*, *Pseudomonas spp.*, *Klebsiella spp.*, *Staphylococcus spp.* and E.coli, this results did not agree with that observed by Jibran and Hadi 1990., Al-Samarai et al 1990 (2, 3), although they reported high incidence of these bacteria in their studies, this difference might be due to the changing of the patterns and ecology of the bacterial infections (5, 11). Also this variation attributed to the difference in the sampling and the type of the wound specimens. The incidence of Coagulase negative staphylococci was reported by many previous authors (2, 3, 12, 13).

Klebiella pneumonae and Pseudomonas aeruginosa which were isolated from both surgical and traumatic wounds showed high percentage of resistance to the different antibiotics, this also formerly reported (11, 12, 14, 15), this attributed to the continuous use of antibiotics and the rapid occurrence of the antibiotic resistance factor in these bacteria (5, 11, 12)> Most of bacterial isolates from both traumatic and surgical wounds showed high resistance against tetracycline this also observed by Maki. (1983) (16), while Hamoudi (17) found most of burn wound bacterial isolates were highly sensitive to tetracycline.

This difference in the results might be attributed to the bacterial strain variation.

The failure of isolation of the other bacteria which were reported by other researchers (2, 3, 13), might be due to the lower dose persistence of these microbes (18) or due to environmental variations (5).

Finally high care and attention must be taken for handling traumatic and surgical wounds to prevent the serious bacterial infections and their antibiotic resistance, through the accurate etiological diagnosis and the proper antibiotic choice. Also the proper disinfection and high precaution must be taken in the surgical wards to decrease the chances of nosocmial wound infections.

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Effects of Post-Irradiation Storage Periods and Temperature on Radiosensitivity of Rice Caryopses *Oryza Sativa* L. Var *Yareet*

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الخلاصة

تم اجراء الدراسات لتحديد علاقة ومعنوية فترات الخزن ودرجة الحرارة بضرر الاشعاع على حبوب الرز ، وقد أدى الخزن الى زيادة ضرر الاشعاع وكانت الزيادة في الضرر أقل عند درجة حرارة ٤ درجة مئوية عنها في درجة حرارة الغرفة (حوالي ٢٥ درجة مئوية) مما أدى الى الاعتقاد بحدوث ترميم حراري لجزء من الضرر . وخلال السنة الاولى من الخزن تحت درجة حرارة الغرفة وجدت زيادة في ضرر الاشعاع ويعتقد ان هذا الضرر مرتبط بسلوك الجذور الحرة طويلة العمر وأوضحت التحليلات الاحصائية للنتائج عدم وجود تحور معنوي في ضرر الخزن بعد انقضاء السنة الاولى ويعزى ثبات الضرر الى أحتمال نضوب الجذور الحرة نتيجة أعادة اقترانها أو اتحادها مع الأوكسجين أو نتيجة حدوث التفاعلين معاً . وأشارت النتائج الى أن زراعة الحبوب يجب أن تتم بعد تشعيعها مباشرة لتقليل الضرر الناجم عن الخزن بعد الأشعاع .

ABSTRACT

Studies were carrie
irradiation storage periods and temperature to radiation injury induced in rice caryopses. Post-irradiation storage increased radiation damage, this increase was less at 4°C than at 25 °C . It is suggested that low post-irradiation storage temperature could thermorestorate part of storage damage . During the first year of storage under room temperature (about 25 °C) there was an increase in radiation damage . It is presumed that the pattern of this damage could be correlated to the behaviour of long-lived free radicals. Statistical analysis of the results showed that long period of storage (more than one year) did not appear to modify post-irradiation storage injury. The stability in damage was probably caused by the decay in free radicals due to radicals recombinations and/or to oxygen-radical combination . The results indicated also that sowing of rice caryopses must take place as soon as possible after irradiation to minimize storage effect.

INTRODUCTION

The effects of irradiation depond on a series of factors. It depends not only on various properties of the material irradiated, but also on factors operating during and after irradiation (1). The conditions in which the material is stored after irradiation and the storage periods are of the most important factors that play a role in increasing, decreasing or mediating radiation injury (2, 3, 4).

Although a major role of seed water content is to mediate the extent of oxygen effect, the results obtained by Constantin et al. (5) suggested that, the increase in water content from 2.7 to 13 per cent afford a degree of protection which is oxygen independent. They also

which is oxygen independent. They also indicated that storage effects were observed during as well as after irradiation in seeds with less than 13 per cent water when the oxygen effect was eliminated. Another investigators demonstrated that the injury to barley seeds from a given dose of x-rays can be enhanced by post-irradiation storage (6, 7), by contrast Justice and Kulik (8) found that as post-irradiation storage time increased sensitivity to oxygen decreased and recovery occurred.

The present experiments were carried out to determine the effect of gamma irradiation in dependence on the post-irradiation storage periods and temperature.

MATERIALS AND METHODS

Caryopses of *Oryza Sativa* L. var. Yareet had been screened for size uniformity and the moisture content of the caryopses was equiliberated to 15 per cent. These caryopses packaged in polyethelene, received 0, 15, 20, 25 and 30 KR. of ^{60}Co gamma radiation at 0.3342 KR./sec. After irradiation, caryopses were planted, immediately as previously described (9) or stored (in their polyethelene packages to keep their moisture content constant) either in refrigerator (4 °C) for 8 days or at room temperature (about 25 °C) for different length of time (8, 181, 372, or 722 days) before planting.

The irradiation times were so arranged that sowing of caryopses of different storage time as well as of those used as corresponding controls could be done at one time. All treatments were replicated four folds using 12 caryopses per replication.

Statistical analysis using blocks method (10) were also done. Reduction in seedling height recorded after 30 days, was taken as a criterion of radiation injury and post-irradiation damage (11, 12).

RESULTS AND DISCUSSION

The effect of post-irradiation temperature

Tables 1 and 2 demonstrate that for a short period of post-irradiation storage (8 days) in both 25 °C and 4 °C seedling heght decreased with much significant reduction occurring at 25 °C.

It seems that under low post-irradiation temperature part of gamma-induced storage damage could be repaired leading to suppression of injury progression in rice caryopses. That means 8 days storage at 4 °C resulted in a distinct thermorestoration of post-irradiation storage injury.

The present observation are different from those of Solar (13) and Byonove et al. (14). The differences apparently due to some unemphasised conditions such as caryopses humidity pre-irradiation, dose rates and varietal difference.

Table 1 : Influence of post-irradia -tion storage temperature on the sum of seedling height developed from rice cryopses (8 days storage).

Radiation dose (KR)	Seedling height in centimeters		
	non-stored caryopses	stored caryopses at 4 °C	stored caryopses at 25 °C+1 °C
0	547	544	549
15	437.6	380.8	302
20	295.4	244.8	109.2
25	240.7	185	109.8
30	191.5	149.6	71.4

Table 2 : Analyses of variance for Table 1.

Source of Variation	Sum of squares	Degree of freedom	Variance	Calculated Value of F	Critical value of F	
					5%	1%
Total	67162.18	59				
Block	4.98	3	1.66	0.40	2.83	4.29
Treatments	66982.43	14	4784.46	1150.11	1.94	2.54
Error	174.77	42	4.16			

C.V. = 3.2%

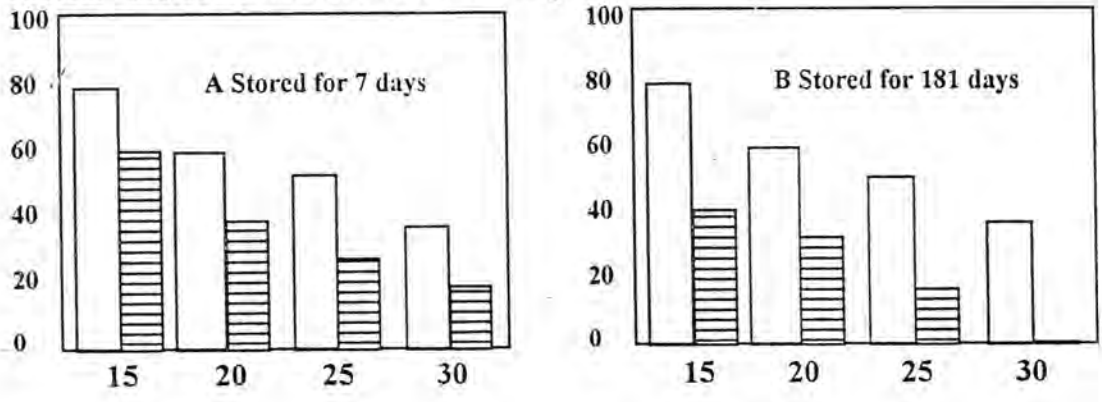
Nevertheless our results concur with those of Howard et al (15) indicating that low temperature reduces the rate of respiration and consequently decreases post-irradia -tion storage effects (16).

The Effect of post-irradiation-storage periods .

Figure 1 and Table 3 show that as post-irridation storage time increased. Seedling height dramatic -ally decreased. The progression of this damage was more obvious in seedlings grown from caryopses irradiated with high doses (25 and 30 KR). In contrast the reduction in seedling height developed from irradiation non srored caryopses was much less. In general there was no modification of post-irraiation storage injury after the first year of storage. The observed reduction is in agreement with previous findings in barley and rice (11, 13, 14).

Two explanations for this considerable damage in the sotred but not immediately grown caryopses might be possible. It must be some factors which are operating after, but only slightly or not at all during irradiation (12). It is clear that the free radicals present post-irradiation engage in a long series of radical molecule reactions which might lead to development of storage damage (17). The second explanation is that : Caropses moisture actually influences the interaction of oxygen and free radicals (forming peroxyl radicals) produced in the seed by radiation. Howeverif the radical-radical interacti -ons results in inct -ivation and radical- oxygen reactions results in damage (due to formation of harmful molecules formed from peroxyl radicals), concurrent action of the two mechanisms could account for the variable damage occurring in storage (18). Thus the pattern of damage can be correlated to the behaviour of long-lived radiation-induced radicals (19). In the other hand planting caryopses immediately after irradiation might act by increasing mobility of free radicals and facilitating recombination to form relatively harmless products before peroxyda -tion can occur.

Presumably after long period of storage (more than one year) there were a rapid decay in free radicals due to radicals recombination or oxygen-radicals combination (20). Thus the stability of damage after one year post-irradiation storage is probably correlated with the lifetime of free radicals resulted in caryopses afte- irradiation.



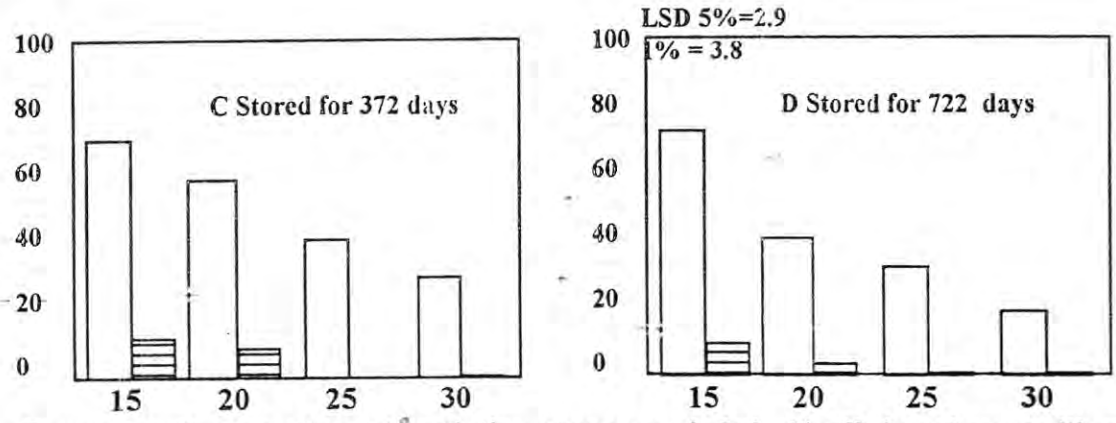


Figure 1 Influence of post-irradiation storage periods in % off cintrol on seedling height developed from irradiated non stored ☐ or irriated stored ▤ rice caryopses

Table 2 : Analyses of variance for Table 1.

Source of Variation	Sum of squares	Degree of freedom	Variance	Calculated value of F	Critical value of F 5%	1%
Total	152830	79				
Vlock	0.7	3	0.23	0.08	2.79	4.20
Treatment	152667	19	8035.11	2819.34	1.85	2.39
Error	163.3	57	2.85			

C.V. = 4.56%

LSD 5% = 2.4
1% = 3.2

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Field and Laboratory Studies on The Effect of Storage Remerathures on Speed and Seed Germination of Tomato Seeds *(Lycopersicon Esulentum).*

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الخلاصة

تم اختبار تأثير درجات حرارة الخزن المختلفة لبذور نبات الطماطة صنف (سوبر مريموند) باستخدام تصميم تام التعشية (CRD) وتصميم القطاعات العشوائية الكامل (RCBD) على نسبة انبات البذور وسرعة انباتها تحت ظروف المختبر والظروف الحقلية على التوالي . اشارت النتائج بأن أعلى نسبة انبات وسرعة انبات تم الحصول عليها في البذور المخزونة تحت درجات حرارة ١٠م° وفي الظروف المختبرية والحقلية بينما أقل نسبة انبات حصلت في البذور المخزونة تحت درجات حرارة ٤٠م° اضافة الى وجود اختلافات بين القراءات الحقلية والمختبرية .

ABSTRACT

Tomato seeds *Lycopersicon esculentum,* Supper marmond cultiver were subjected to different storage temperatures; 0, 10, 20, 30 and 40 C° for two months. Their germination percent and speed of germination germination were studied under field and laborotary conditions. Highest speed and seed germination were occurred at 10 C°. The lowest were at 40 C°. Dissimilarties between field and laboratory trails were noticed. Counting for seedling emergance should not exceeds more than 9 days.

INTRODUCTION

It has been generally accepted that seed germination is a critical stage in the life cycle of a plant. And is considred by some investigatores (2,5) as the road seeds must travel before it is becoming a seedling and consequently a mature plant. Since early plant estibishment offers the potential for earlier crop harvest, and delayed germination and seedling emergence extend the period form planting to crop harvest, satisfactory method can be used for some crops to reduce the time necessary for seed germination and seedling emergence to achieve earlier crop harvest. Several investigators (1, 2, 6) reported that germination of most seed types is governed by the balance between seed germination inhbitors and promoters. Amen (1) stated that when growth inhibitors are found in a physiologically greater a mount than promoters, seed germination is blocked. While the reverse condition triggers the germination process. Exposing the seeds to an agent that would alter the balance between seed inhibitors and seed promoters might be used to activate or control seed germination process. A trigger agents such as exposing seeds to light to activate germination process of several aeed types are available in the literatures. However, little is known about the effect of storage temperature on seed germination and speed of germination (vigour) of tomato seeds.

The work reported here is to determine the effect of different storage temperatures on seed germination and speed of germination of tomato seeds, Supper Marmond cultivar in a field and laboratory study.

MATERIALS AND METHODS

Bulk amounts of tomato seeds Supper Marmond cultivar were placed at 0, 10, 20, 30, and 40 C° storage temperatures for two months. The seeds were bought from the local market at the year in which the experiment was held (1991) and claimed to be as a product of 1990. The experiment thus, involved five treatments. Twenty seeds of each of the temperature subjected bulk seeds, were taken and placed in a Petri-dish size (6 x 1cm). All Petridishes the, placed in the laboratory (at 25 C°) for germination test. Each dish was considred as one experimental unit. Six replications per treatment were used and analyzed as compeletly randomized design (CRD). The same procedure was followed for the field experiment except seeds were placed in pots size (20 x 20 cm) instead of petri-dishes.

The pots were then placed in a sunny well aerated fiel at 21st of March. Data on seed germination for the laboratory and field experiments were taken three and four days after sowing, respectively. Seeds were considered germinated when primary shoots has appeared and roots were about 2 to 3 mm long. Speed of germination was determined by using the vigour index formula as described by Camargo and Vaughan (4) :

$$VI = \sum NX/DX$$

where NX = number of seeds germinated on x days and DX = number of days from the beginning of germination test to x day. The same formula was utilized to determine the vigor index criterion of the laboratory and the field grown tomato seeds. The high values of vigour index indicates higher speed of germination and the lower values for the low speed of germination. The design of the field experiment was Randomized Compelete Block design (RCBD). When significant "F" values were obtained for the treatment effect, the Duncun multiple range test at 0.01 level of significance was used to compare treatments means.

RESULTS

Laboratory experiment

Results of germination percentages as well as the speed of germination as indicated by the vigour index (VI) are shown in table (1). This table showed that the highest germination percentage was occurred when seeds were stored at 10 C°. And the lowest was at 40 C°. No significant differences in seed germination percentage were found when seeds were stored at 0 C° compared with 20 C° and at 20 compared with 30 C°. The highest speed of germination was at 10 C° and the lowest at 40 C°.

Although there were no significant differences in speed germination of seed stored at 0.0, 20 and 30 C°, the former tended to have higher vigour index than those of 20 and 30 C°.

Field experiment

Ruselts of germination percentage as well as speed of germination (VI) are shown in table (2). It can seen from this table that the highest percent of germination and speed of germination were obtained when seeds were stored at 10 C°. Also percent of germination was decreased when storage temperture was below 10 C°, (i.e at o C°). However when storage temperature was higher than 10 C° highest reduction occurred when storage temperature was to 40 C°. No significant differences were found in

germination percentage between seeds stored at 20 and 30 C°, however their speed of germination were significantly different (at 0.01 level of significance).

Table 1 : Effect of storge temperatures on germianation percentage and speed of germination of tomato seeds grown in the labratary

Storge Temperatures	Germianation Percentage	Vigor Index
0.0	70.83 b[Z]	1.59 b
10	81.66 a	1.87 a
20	65.0 bcd	1.44 bc
30	59.16 d	1.31 bcd
40	47.50 e	1.05 e

Table 2 : Effect of storge temperatures on germinanation percertage and speed of germination of tomato seeds grown in the field

Storge Temperatures	Germianation Percentage	Vigor Index
0.0	48.33 b	1.07 b
10	58.30 a	1.29 a
20	40.83 c	0.90 c
30	35.00 cd	0.77 d
40	6.67 e	0.15 e

Z Mean separation within columns by Duncan multiple range test, 1% level.

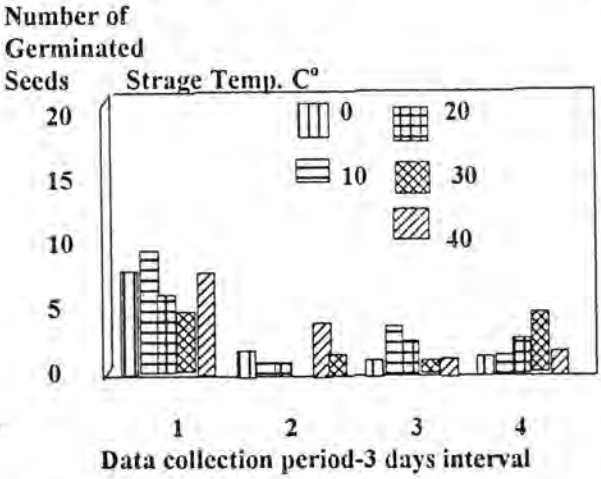

Figure 1: Number of seeds emerged at each counting period in the labratory for all temperatures included in the experiment.

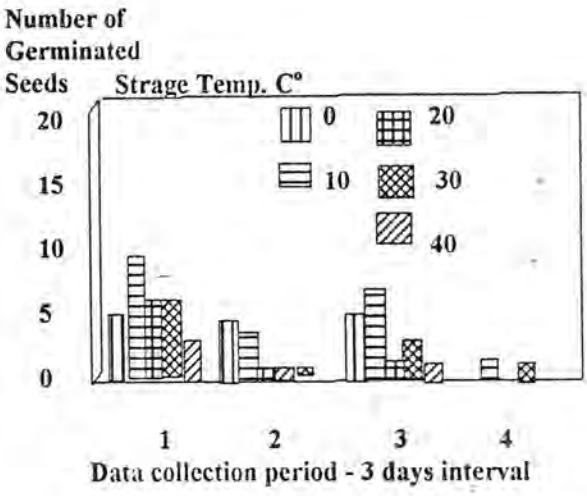

Figure 2: Number of seeds emerged at each counting period for all temperatures included in the field experiment.

The highest number of germinating seeds, of all storage temperature, were occured at the first counting period for both laboratory and field experiment Fig (1,2).

DISCUSSION

It is clear from this experiment that storage temperature has a great influence on both seed germination and speed of germination (VI) of tomato seeds, Supper Marmond cultivar. Temperature as low as 0 C° or higher than 10 C° altered not only seed germination percentage but also the speed at which germination occurred. The highest germination percentage and speed of germination that occurred at 10 C° for both laboratory and fueld experiment suggest that the best storage temperature to store tomato seeds is at 10 C°. This is to ascertain not only higher germination percent but also higher germination speed.

Because subjecting seeds to 10 C° achieved a rapid shoots and radical protrution from the seed coat. And as a result, the time necessary for seedling emergance is reduced. This offers the potential for better stand development and earlier crop harvest. These results

agreed with (1, 7). The dissimilarities between the field and the labratory trail is expected due to the fact that all conditions at the labratory experiment were under control. The reduction in the seed germination at low temperature (0.0 C°) may be due to the indirect effect of this low temperature on the viability of the embryo of the seed. It is possible that too long period of prechilling at this temperature (two months in this experiment) caused a weakened and or deformed embryo, and as result seed and speed of germination were lowered. The highest speed and seed germination at 10 C may be due to the nature (1) and the activity of the protolytic enzymes (6) which was enhenced at this temperature. This enzyem in conjection with cellulase serve tp degrade cell wall (1). The latter is considred as an essential step in the loosening of seed coat prior to radical protrution and for this reason seedlings emergence were delayed. The reduction in seed germination and speed of germination at higher temperture may be due to the influnce of high temperature on seed respiration during storage period.

Because, seeds as a biological units would increase the rate of their biological processes such as respiration at high temperatures. This increase in respiration places a demand for carbohydrates, protein and other food reserve material neccessary for germination and thus the latter was reduced or compeletly ceased. It also seemed from this experiment that extending the counting periods up to 15 days did not contribute much to the total emerged seadlings regardless of storage temperature.

And for this reson counting for seedling emergance should not exceeds more than 9 days after sowing.

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Changeable Bacterial Flora in Burn Wound Infection During Hospitalization

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الخلاصة

التغير الحاصل في انواع الجراثيم المعزولة من خمجات الحروق خلال فترة البقاء في المستشفى . تم في هذا البحث متابعة حالة (٥٠) مريضا راقدين في مستشفى اليرموك التعليمي يعانون من حروق مختلفة الشدة وذلك لفترة اسبوعين ما بعد الحرق لغرض دراسة التغير الحاصل في انواع الجراثيم المعزولة وقد لوحظ ان نسبة تواجد وعزل البكتريا من الحروق تزداد بمرور الوقت وان جرثومة *Staph. aureus* هي السائدة خلال الاسبوع الاول ما بعد الحرق ثم تحل محلها الجراثيم السالبة لصبغة كرام وبالاخص جرثومتي *Ps. aerygunosa* و *K. pneumoniae* .

ABSTRACT

Fifty hospitalized burn patients admitted to AL-Yarmook teaching hospital were followed-up for a period of (2) weeks , during 1992 . Wound cultures are studied to determine the changeable pattern of bacterial flora colonizing burn wounds in relation to time . *Staph.aureus* was commonly isolated in the first week and was later replaced by Gram-negative organisms mainly *Ps.aeruginosa* and *K.pneumoniae* . The colonization rate by burns increased with increasing duration of hospital stay .

INTRODUCTION

Infection is an important cause of morbidity and mortality in burns (Pruitt,1984) , the changing pattern of bacterial species with time is a prominent feature related to the colonization trend (Hansbrough et al. , 1985).

Differences in species and colonization rate particularly within the first week postburn were reported (Yemul and Sengupta,1981, Sawhney et al.,1989). The changes apparently represent part of the normally colonization process which leads to the establishment of the more permanent flora . The type of organisms prevailing in the environment state of burn patient , nature of the burn wound and its anatomic location, all can effect such changes (Brook and Radolph, 1981).

In this study the effect of time on the microbial colonization pattern of burn wounds was assessed .

PATIENTS AND METHODS

Fifty patients with different degrees of burn , ranging between 10-80% BSA , were followed-up over two weeks period of hospitalization . 300 burn wound specimens were collected , six specimens from each patient taken on the 1st. , 3rd. , 5th. , 7th. , 10th. and 14th. days postburn .

The wounds samples were collected with sterile cotton swabs , cultivated on : MacConky , Blood and nutrient agar , incubated at 37°C for 24 hrs. , and the

organisms were identified morphologically and biochemically using the API 20E system for Gram-negative bacilli , and the routine conventional procedures for Gram-positive bacteria (Fine-gold and Baron,1986).

RESULTS

A total of 350 bacterial isolates which belonged to six species were isolated over two week period of follow-up (Table-1) .

Ps.aeruginosa was the most common (44%) followed by *K.pneumoniae* (30.2%) and *Staph.aureus* (24%) , (Table-1-).

The bacterial colonization rate showed in increase with the passage of time (Table-2). The rate was 28% , 60% , 90% , and 100% on the 1st. , 3rd. ,5th. and 7th. days postburn , respectively.

Table-3 revealed that both *Ps.aeuginosa* and the coliforms had the lowest isolation rate on the 1st. day post burn (1.3% and 3.6% respectively) , then Ps.aeruginosa showed an increase in rate with time , reaching a peak by the end of the second week (29.9%) , where as the coliforms organisms showed a peak of isolation rate 7 days postburn , after which gradual decline occurred the second week . *Staph Aureus* had a completely different pattern , as the isolation rate was high on the first day post burn , peaked 5 days postburn , after which a sharp decline was noticeable towards the end of the first week .

Table (1) : Bacterial profiles of 350 isolates recovered from 50 hospitalized patients with infected burn wounds during 14 days follow-up

Species	Pure culture number &%	Mixed culture number &%	Total number &%
Ps. aeruginosa	77(22%)	77(22%)	154(44%)
Coliforms	27(7.7%)	83(23.7%)	110(31.4%)
Staph. aureus	36(10.3%)	48(13.7%)	84(24%)
Total	140(40%)	210(60%)	350(100%)

Table (2) : Results of burn wound cultures from 50 hospitalized patients during 14 days follow-up

Cultures	Days postburn						
	1st.	3rd.	5th.	7th.	10th.	14th.	Total
Sterile	36(72%)	20(40%)	5(10%)	0	0	0	61
Single isolate	12(24%)	19(38%)	23(46%)	27(54%)	27(54%)	33(66%)	140
Multiple isolate	2(4%)	11(22%)	22(44%)	23(46%)	23(46%)	17(34%)	99
Total	50	50	50	50	50	50	300

Table (3) : Changeable pattern of bacterial flora in 50 hospitalized burned patients during 14 days follow-up

Bacterial Species	Days Postburn						
	1st.	3rd.	5th.	7th.	10th.	14th.	Total
Ps.aeruginosa	2	6	22	34	44	46	154
Coliforms*	4	17	22	26	24	17	110
Staph.Aureus	10	20	27	15	8	4	84
P.mirabilis	0	0	0	1	1	0	2
Total	16	43	71	76	77	67	350

* Coliforms K.pneumoniae , E.Cloacae and E.coli .

P.mirabilis appeared by the end of the first week and disappeared towards the end of the second week and this may point out the hospital as a probable source of infection .

DISCUSSION

Infection is a major challenge in the treatment of burns . The spectrum of infective organisms varies according to the geographical area and from time to time (Yemul and Sengupta , 1981).

The finding of *Ps.aeruginosa* in this study with such a high ranking order was not unexpected and it is predominance in burn patients cast peculiarity on the trend of burn wound infection .

A unique feature demonstrated in this study was the early colonization of burn wounds (28% on the first day) , probably of certain bacteria to proliferate rapidly after surviving the initial trauma , taking the advantage of absent local defenses during this time (Deitch et al. , 1985) . The increasing rate of colonization with time suggests an environmental source of bacterial colonization (Ramakrishnanet al. ,1985 ; Sawhney et al. , 1989).

One of the peculiar features of burn wound infections is that the microbial pattern is constantly changing (Bharadwaj et al. , 1983) and results obtained in this revealed that Gram-positive organisms showed initial predominance in the immediate postburn supplanted by Gram-negative flora which gains ascedancy in the later part of the first week and remained predominant till the end of the second week postburn.

the fluctuation in the colonization trend of burn wound microbial flora is probably demonstrating an environmental selective pressure which determine a balanced association between the colonizers , and the outcome would be the establishment of the permanent microbial flora (Jawetz , 1987). However , the type of bacterial first colonizing the wound is dependent on treatment and the environment to which the patient is exposed (Dimick , 1984).

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The influence of Liguorice Waste Materials on Nitrate, Soil Structure and Barely Growth

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الخلاصة

مخلفات معمل عرق السوس المتبقية بعد استخلاص مادة عرق السوس من جذور نبات Glycyrrhize glabra في معمل العزيزية ، اضيفت الى التربة الغرينية الطينية المزيجية المأخوذة من منطقة الهارثة في البصرة بنسبة ١ ، ٢ ، ٣ ٪ على اساس الوزن الجاف في تجربتين احداهما في المختبر والاخرى في البيت الزجاجي لدراسة تأثير هذه المادة على النترات المتكونة وتركيب التربة ونمو الشعير . ان المخلفات المضافة الى التربة ادت الى زيادة النترات بصورة واضحة في تجربة المختبر وكذلك لوحظ زيادة في تحيب التربة وقلة في الكثافة الظاهرية للتربة المعاملة . في تجربة البيت الزجاجي تحققت زيادة في نسبة الانبات والنمو للشعير نتيجة لاضافة هذه المخلفات ووجد ام معامل الارتباط بين نمو الشعير في تجربة البيت الزجاجي وتجمع النترات في تجربة المختبر معنوياً عند احتمال ١٪ وقيمته ٨٢٪ .

ABSTRACT

The waste materials that remain after liquorice extraction from the root of Glycyrrhiza glabra were added to a silty clay loam soil at rates of 1, 2, and 3% liquorice waste in laboratory and greenhouse studies to determine the effect on barley (Hordeum vulgare) growth, soil structure, and on nitrate accumulation in soil. Liquorice material added to the soil had drastically influenced the NO_3^--N content in laboratory study. As the rate of liquorice material increased the accumulation of NO_3^--N increased. It was found also that liquorice increased the aggregation of the soil and decreased the bulk density of the treated soil. The addition of this material to the soil in a greenhouse study resulted in an increase in the germination percentage and the growth of barley. The relationship between barley growth in the greenhouse and nitrate accumulation in the laboratory soil had a correlation coefficient of 0.82 (P = 0.01).

INTRODUCTION

Mesopotamian plan is the most important area of irrigation agriculture in Iraq, however, most of these soils are saline, low in organic matter and nitrogen content[1].

Improvement to the fertility of these soils can be made by applying organic matter of chemical fertilizer, many soils produce low yields because the poor soil physical condition.

Source of organic matter should be applied to such soils in order to improve their productivity. Industrial products such as date and sugar beet waste were tested to improve the soils structure of soil taken from the Mesopotamian plain in central Iraq. They were thought to

improve the physical properties of the treated soils[2].

The present investigation was under taken to:

a- determine the best rate of liquorice waste to supply NO_3-N for plant growth.

b- examine the influence of this amendment on soil structure.

The liquorice fiber remaining after liquorice extraction is particularly worthy of study since these plants grow wild producing high yields without any cultivation. The average production of liquorice roots is about 5 to 10 tons per hectare and the annual production is the range of 20000-25000 tons. It is therefore, of great interest to study this material to see the effect on soil properties because it is very cheap and easy to use without special equipment.

MATERIAL AND METHODS

LABORATORY INCUBATION:

The soil sample was taken from top 0-45 cm of El-Hartha silty clay loam soil. During air drying, the soil was gently crumbled by hand and the 2-3 mm fraction was separated out by sieving. Some of the soil properties are shown in Table 1. Air-dried waste material from Azeezea liquorice root extraction factory was ground to pass through 2mm sieve and throughly mixed prior to use. Some properties of liquorice materials are shown in Table 2. The experiment comprised a completely randomized design with five treatments (untreated soil, soil + 1% liquorice, soil + 2% loquorice, soil + 3% liquorice and soil + 0.25% NH_4NO_3) and four incubation periods. 0, 20, 40, and 60 days all with three replications.

The liquorice and NH_4NO_3 were mixed with 200 g of soil and placed in small plastic containers. Water was added to the treated and untreated soil to reach field capacity. Alid was placed on each container and were incubated at room temperature (22-25)°C for the indicated time periods. At the designated time period (60 days) the appropriate samples was extracted with 2NKCl by shaking for 1 hour to determent the nitrate and amonium[3,4].

The wet sieving method was used for assessment of aggregate stabil[5].

The term stability factor (SF) was used and defined as follow:

$$SF = \frac{\text{sum of soil weights retained the 3mm.2mm sieves}}{\text{oven dry sample}} X100$$

Thus the larger the value of the SF the greater the stability of the soil under the test.

Bulk densities of the potted soils were determined on an oven dry weight basis as follow :

$$B = \frac{M}{V}$$

where B= the Bulk density (g/cm^3).

M=oven dry mass of the soil (g).

V=the volume occupied in the pot (cm^3).

GREENHOUSE EXPERIMENT

The same design used in laboratory experiment was used in the greenhouse study. The appropriate treatments were added to 1000 g of the air-dry soil and the mixtures placed in plastic pot. Three seeds of barley were planted. Germination and emergence counts were made 3,6 and 9 days after planting. The plants were grown for a period of 51 days. Harvested plant samples were dried in an oven at 70 °C for 24 hours then weighed to determine dry matter accumulation.

RESULTS AND DISCUSSION

LABORATORY STUDY:

Analysis of variance showed that all main effects and interactions were significant. The treatment means and interacation of ammonium and nitrate are summarized in tables 3 and 4.

Table 3 shows the NH_4^+-N data for liquorice and NH_4NO_3 treatments. Liquorice treated soil showed a rapid increase, then a rapid drop in NH_4^+-N contents during the first 20 days.

The results in Table 4 show the NO_3^--N accumulation value was greatest for 0.25% NH_4NO_3 followed by 3%, 2%, 1% and untreated soil. The highly significant interaction between treatments and time indicated that nitrate formation was dependent upon both treatment and time of incubation.

GREENHOUSE STUDY:

In the greenhouse study, there were no measurable differences in the number of barley seed emerged at 3, 6 or 9 days after seedling in liquorice treated soil. Thus, there was no emergence inhibition. As can be seen, soil treated with liquorice is more aggregate then both, the untreated and the 0.25% NH_4NO_3 treated soil.

Both seed germination and plant growth were retreated in untreated and 0.25% NH_4NO_3 treated soil. This was due to the decrease of mineralization of organic N [6,7,8,9].

Analysis of variance of the data from the greenhouse study indicated that growth of barley was affected by liquorice treatments significant at 5% level. It is clear that with increasing liquorice, the dry matter of barley was significantly increased (Table 6). The limited growth in untreated soil was probably due in part to the lack of nitrogen available to the plants. However, the decrease in plant growth of the 0.25% NH_4NO_3 treated soil probably was due to the increased salt content of this soil. The NO_3^--N accumulation data (Table 4) resulting from various treatments explain some of the differences in barley growth in the greenhouse and NO_3^--N accumulation after 2 months incubation were chosen because the barley plants were grown for about the same time before harvesting.

INTERRELATIONS OF SOIL NITRATE - NITROGEN AND THE GROWTH OF BARLEY

Liquorice material could be added to the soil at optimum rate (2%) with an enhancing effect on barley growth. This effect on barley growth (dry matter accumulation) in the greenhouse was related to NO_3^--N content of the soil. The following regression equation describes this relationship:

barley growth = $0.7588 + 0.061734\ (NO_3^-\text{-N}) - 0.000079\ (NO_3^-\text{-N})^2$

This equation indicates that maximum barley growth under the conditions used in this study was obtained when the treatments were the same as those resulting in about 300 ppm of NO_3^--N accumulation in 60 days (Fig. 1).

Table 1. Some physical and chemical properties of soil used in study:

Coarse sand	(2000-200μm)%	6.8
Fine sand	(200-20μm) %	8.2
Silt	(20-2μm) %	55.0
Clay	(< 2μm)%	30.0
Total		100.0

Texture silty clay loam

Field capacity %	20.5
C%	0.57
N%	0.07
C/N	8.14
O.M	0.9
$CaCO_3$ %	2.8
$CaSO_4$ %	0.51
Ec 1:1 ms. cm^{-1}	5.1
pH	7.5

Table 2. Some chemical properties of liquorice material:

C%	32.3
O.M %	56.2
N %	1.9
P %	0.26
C/N %	17
pH	8.0
EC 1:1 ms. cm^{-1}	1.5

Table 3. Effect of liquorice and NH_4NO_3 on NH_4^+-N accumulation in the soil laboratory

Soil treatment	NH_4^+-N ,ppm Length of incubation (days) 0	20	40	60	Average of treat-ment	LSD 5%
Untreated	14.8	12.8	8.5	4.9	10.0	
1% Liquorice	88.8	78.4	67.0	30.8	66.2	51.3
2% liqurice	101.3	79.0	68.7	48.3	74.4	
3% liquorice	162.0	81.0	71.7	56.0	92.7	
0.25% NH_4NO_3	120.6	1	2	163.8	107.6	
Average of incubation (mean)	90.4	71.2	63.2	49.9	interacation 5% LSD	
5% LSD	19.1				24.7	

Table 4. Effect of Liquorice and NH_3NO_3 on NO_3^--N accumilation in the soil in laboratory:

Soil treatment	NO_3^--N ,ppm Length of incubation (days) 0	20	40	60	Average of treatment	LSD 5%
Untreated	16.2	20.7	25.4	27.3	22.4	
1 % Liquorice	38.4	75.4	98.4	163.6	39.9	69.3
2 % Liquorice	52.1	85.7	99.2	233.2	117.6	
3 % Liquorice	61.7	89.6	103.0	373.1	156.9	
0.25 % Liquorice	33.0	40.4	500.7	599.5	458.6	
Average of incubation (men)	99.7	135.1	165.3	279.8	interaction 5% LSD	
5% LSD	30.0				21.7	

Table 5: Mean percentage of stability factors (SF) and bulk density of the soil at the end of the incubation experiment:

Soil treatment	aggregate size (mm) >3	3-2	total	bulk density g/cm^3
Untreated	24.3	3.9	28.2	1.3
1% Liquorice	31.4	5.2	36.5	1.1
2% Liquorice	52.0	5.4	57.4	1.0
3% Liquorice	46.9	7.5	54.4	0.9
0.25% Liquorice	21.5	4.3	25.1	1.4
LSD (5%)	6.6	1.00	7.6	0.1

Table 6. Effect of Liquorice and NH_4NO_3 on barley seed germination % and dry weight as a measure of growth (g/pot):

Treatment	*Barley seed germination %			Sry weight of barley Plant/pot
	3 days	6 days	9 days	
Untreated	.....	33.3	55.6	2.4
1% Liquorice	100	100	100	9.2
2 % Liquorice	100	100	100	12.0
3 % Liquorice	100	100	100	12.9
0.25 % Liquorice	.....	77.8	77.8	7.8
				LSD 5%

* These results are not included in the statistical analysi

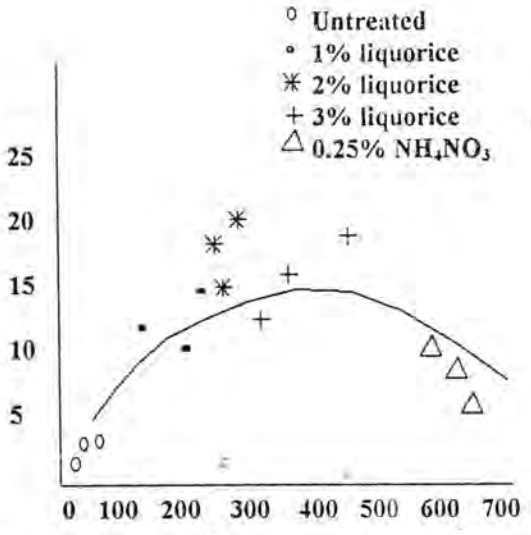

Figure 1: Correlation between the 60-days barley growth in the soil in greenhouse and NO_3^--N accumulation in the same soil in the laboratory.

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Study of Link Turbidity Factor and The Influence of Precipitable Water Vapour in Baghdad

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الخلاصة

أجريت القياسات الساعية لقيم الاشعاع الشمسي العمودي المباشر بأستخدام جهاز ايلي (NTP) في مدينة بغداد . سجلت اكثر من ٢٦٠٠ مجموعة من الوحدات الساعية والتي أجريت للأيام الصاحية الخالية من الغيوم للفترة من كانون الثاني ١٩٩٢ ولغاية كانون الاول ١٩٩٣ . النتائج تشير الى ان أعلى قيم لعامل عكره لنك سجلت خلال أشهر الصيف (الحاره والجافة) واقل قيم لها سجلت خلال أشهر السنة (الباردة والرطبة) . مما يتوافق هذه النتائج مع كثير من الدراسات التي أجريت في دول أخرى من العالم . كما أجريت دراسة تحليلية لقيم معامل عكره لنك عند حدود معينة متعارف عليها فوجدت أن قيم تشكل أكثر من ٤٠٪ من الرصدات الساعية عند القيم 4<T<5 بينما في الحالات التي يكون فيها الجو عكره جدا تشكل ٧٪ فقط من الرصدات الساعية . كما تم دراسة تأثير بخار الماء القابل للترسيب على تغيرات عامل عكره لنك .

ABSTRACT

Hourly measurements of irradiance total direct normal solar radiation were used to calculate the link turbidity factor. More than 2600 sets of observations were chosen under clear sky and cloudless condition during the period of Jan 1992 - Dec 1993. The results showed that high turbidity values recorded in the summer season and lower values observed during the winter. Analysis of the frequency distribution of link turbidty factor was carried out at specified ranges. The results also indicated that more than 40% of the measurement ranged between 4<T<5, while turbid cases represent 7% only. The influence of the perceptible water content and atmospheric turbidity factor at noon were analyzed also.

INTRODUCTION

The intensity of the extraterrial solar radiation traversing through the earth's atmosphere is attenuated by various constituents of the atmosphere, namely, air molecules, liquid, aerosols, and precipitation in clouds. Theoretical analysis of attenuation of solar radiation passing through clouds requires great deal of information regarding instantaneous thickness, position and number of layers of clouds, as well as their optical properties.

However for technical utilization of solar energy, study of solar radiation under cloudless skies is very important, particularly for solar system performance using concentrators [1].

The cloudless sky generally produce the maximum energy available, this maximum establishes the oriteria with which engineers can design equipment for the utilization of solar energy in the field of solar cells, space heating process, water heating and the thermal loading on buildings. The maximum is also necessary in the design of materials for solar application to prevent early damage or deterioration of plastics.

The ratio of the prevailing attenuation of solar radiation through a real atmosphere versus that through a clean dry atmosphere gives an indication of atmospheric turbidity. Study of atmospheric turbidity is very important in meteorology, climatology and for monitoring of atmospheric pollution; Atmospheric turbidity is rather complex factor as it, depends on air mass, season of the year, latitude and the local atmospheric conditions; it's value must therefore, shows a marked variation; with time and place with a normal annual variation having its maximum value in summer and its minimum in winter [2]. However, the greatest change are those which arise from atmospheric pollution.

LINK TURBIDITY FACTOR T

The first rational approach to the problem was perhaps due to Link and Boda, eventually named Link turbidity factor T, which indicates the number of clean dry atmosphere that would be necessary to produce the same attenuation of the extraterrestrial solar radiation that is produce by the real atmosphere. Its value can vary from 1 to 10, and can't be less than 1.

Link's turbidity factor is a measure of water vapor and dust content of atmo phere, and can be determined with the only measurement of the direct normal solar irradiance as follows :

$$I = (Io/S) \exp(-a.m) \quad \ldots\ldots (1)$$

where Io is the solar constant (1367 w/m^2), and S is the correction factor for mean Sun-earth variation; I is the direct normal solar irradince over all wavelengths received at the earth's surface, and a is the mean extinction coefficient, Hence:

$$T = [(1/a.m) \ln (Io/I.s)] \quad \ldots\ldots (2)$$

where a being the mean Rayleigh extinction coefficient and obtained by Feussner and Dubois, and presented by Kasten [3], through the following simple formula:

$$a = (9.4 + 0.9\ m)^{-1} \quad \ldots\ldots (3)$$

where m is a parameter of the altitude angle.

For standard pressure, accurate values of the optical air mass can be obtained according to Kasten formula as follows [4] :

$$m=[\sin h+0.15(h+3.885)^{-1.253}]^{-1} \quad \ldots(4)$$

where h is the solar altitude angle in degrees.

RESULTS AND DISCUSSION

During the period of research work, January 1992 to December 1993, more than 2600 sets of measurements were taken under clear and cloudless sky conditions, these are instantaneous measurement taken each hour from 8.AM to 6.PM, in the research field observatory of the solar energy research center, Baghdad - Iraq, (Latitude 32 21 N, Longitude 44 25 K, Elevation = 34m above MSL). The site can be regarded as semi - rural region.

The average values of T and standard deviation are presented in figure (1). The figure indicates large monthly variations of T with a distinguished peak in (May - July), because dust storm are known to occur more frequently during these months, while a pronounced low values in January and February.

The annual and seasonal means of Link's factor with their standard deviation c are presented in table (1), where is also indicated in brackets the increase in percent with respect to winter values. It seems that autumn and spring values are rather higher and near to summer values than to winter values. This result agreed with many other studies [2, 6, 7], that indicate maximum T in summer and minimum in winter. For a clearer picture of atmospheric turbidity the relative frequency distribution of Link's factor T is investigated and drawn in figure (2) which represent the distribution for the hourly date of 1992 - 1993 Linked together, this figure seems to reveal a gaussian distribution law and the peaks of the histograms remain around an interval $4<T<5$. The data in this range represents 40%, while in the range $T<4$ represents 25.5% which classified as clear days while in the range $T>6$ represents 7.4% as turbid and humid days.

The monthly average values of Link turbidity factor for different hour of the day were also studied during the period of work, the results are tabulated in table (2), which shows a steady increase from sunrise to reach their highest values at solar noon, then decrease gradually in the afternoon and attain their minimum values at sunset - However, this table is useful in the design of solar energy utilization systems.

Influence of Perceptible Water Content and Atmospheric Turbidity Analysis of Values at Noon

The total amount of water vapor in the vertical direction is highly variable and depends on the instantaneous local conditions, however, this amount generally expressed as perceptible water thickness, and it can be readily computed through a number of standard routine atmospheric observations, such as relative humidity and the ambient temperature (To). Perceptible water content of the atmosphere, W. has been calculated at noon only for the same period of (t) by using Leckner's formula, which express perceptible water in terms of temperature and relative humidity as follows [5] :

$$Wcm=0.493*Q/To \exp(26.23-5416/T) \quad (5)$$

where To is air temperature in degree Kelvin and Q is relative humidity in fraction of one.

The monthly mean values of m, W and T at noon are presented in figure (3), the values of W ranged from 1cm in winter (when m has high values) to 2.3cm in summer (where m has low values). It shows that the turbidity values are highest in May and lowest in November while the perceptible water vapor is highest during the summer seasons.

Using the measured data, the following correlation between T and W has been established for noon time:

$$T = 4.05 + 0.45\,W \quad \ldots\ldots (6)$$

with correlation $r = 0.49$. This relatively low correlation between T and W needs further investigation.

Figure (4) shows a comparison of this study with some others carried out at Halwan, Egypt 1975 [2] and at Montfavet, France 1979 [6]. It is shown that these curves has the same trend with maximum values occur in summer months and minimum values in winter. A pronounced peak of T in April for Baghdad location due to the occurrence of dust storms during this period of the year, it is also shown that the average values of T are higher in Halwan throughout all months of the year, this is because of the industrialization of the Halwan atmospheric environment.

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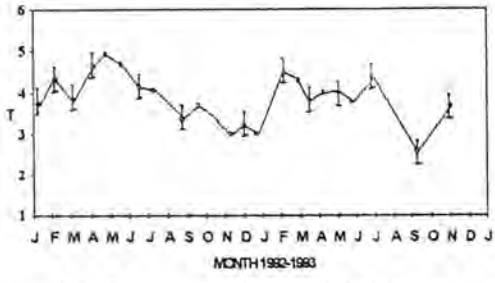

Figure 1: Average values of T and standard deviation

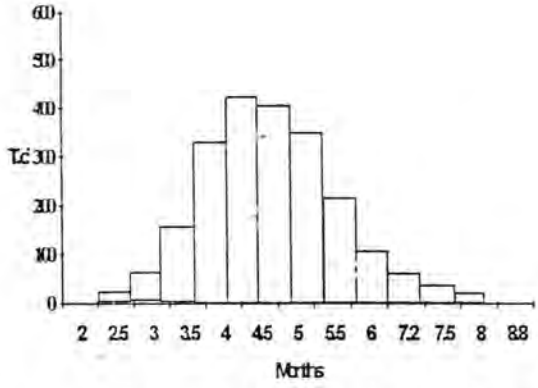

Figure 2: The distribution of link's factor

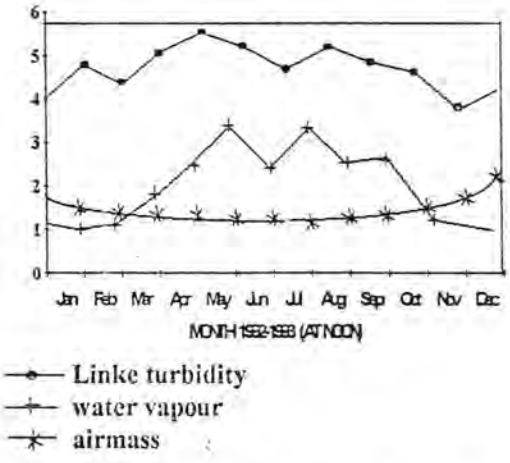

Figure 3: Monthly mean values of m, W and T at noon.

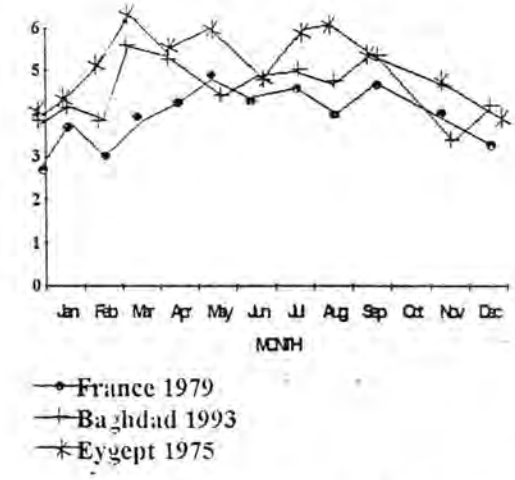

Figure 4: Comparison of T in different places

The Use of Europium Oxide as a Safty Control Rods in Nuclear Reactor

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الخلاصة

يتضمن البحث على الحسابات النظرية بأستخدام (KENO - 4) والذي يعتمد على المجاميع المتعددة التي تعتمد عليها طريقة مونتي كارلو . أن البرنامج المذكور يعتمد على النظرية الانتقالية للنيوترونات . وقد تم في هذا البحث حساب الفيض النيوتروني لستة عشره مجموعة من الطاقات على شرط أن قضبان السيطرة لاوكسيد اليوربيوم (EU_2O_3) الموجودة في مركز المفاعل . يمكن النتائج المستخلة من هذا البحث اعتماد كدليل للدراسات اللاحقة وذلك بأستخدام ممتصات أخرى والممتصات الخليطية للمفاعلات السريعة .

ABSTRACT

This work concern a theor NO-4 which is a multi-group Monte-Carlo program. This program depends on the neutron transport theory. In this work we compute the flux of neutrons for sixteen energy groups in the condition that the control rods of EU_2O_3 were exist in the center of the reactor core. The results which obtained in this work can be used as a basic reference for additional work with other absorbers and also a mixtures of absorbers in the fast training reactors.

INTRODUCTION

The main condition to choose the type of the material used in the nuclear reactors which mainly depends on the reaction range of the neutrons in the material, in other word of EU_2O_3 have a large absorption cross-section for neutrons in the range of thermal neutrons is 8740 barn (1).

Under this condition the EU_2O_3 in the center of the nuclear reaction at a certain amount of masses can satisfy this type of work. Therefore, the results will be effect on two mainly points, firstly the large depression of the neutron flux, secondly the reactivity will change possibly in many states. For this reason mentioned above the reactor cannot work on its designed power.

The natural Europium (Eu) have two isotopes Eu-151 and Eu-153 with percentage 47.8% and 52.2% respectively (2). The Europium have a large cross-section specially in the middle and high energy range, so that in the last few years many worker concentrate on the fast reactor.

In this present work the measurements indicate the characteristic of (EU_2O_3) and Boron Carbonide approximately equivalent specially in the variation of reactivity. Add to above the Europium more suitable then "B4C" in "FTR" as control rods (3).

When the Eu_2O_3 irradiated by the neutrons beam the product will be a chain of radiative nuclides. This type of nuclides leads to have a large cross-section, its value was (430 barn) (3). In the other hand for a natural matter will be reduced to (700 barn) and stay relatively constant in the high density of irradiation.

of theoretically assumption that the Eu_2O_3 rods in the centered, in the neutron trap of (IRT-5000).

Under the condition that we used a different radius of rods the Monte-Carlo Code KENO-4 which represented a simple types of three-dimensional configuration, which depends upon a neutron transport theory.

This Code of program help us to calculate K-effective, life time, generation time, leakages, absorption, neutron flux and fission densities through the reactor composition (5).

CALCULATIONS

1- Radial distribution of neutron flux

In order to simplify our calculation, simply we choose a several different radius have been used from (0.1) to (0.5) cm, we assume that rods of absorber in the cell (28) as represented in Fig. (1) which represented the nuclear reactor core assembly for (IRT).

The radius of the used rods has been varied with a constant hight.

The atomic number density for Eu_2O_3 which used for those homogenized absorbers rods which represented in table (1).

The crosssections and group- dependent data which is used in this code of program are not limited to Hansen Roach cross-section or a sixteen groups. Also this code include. The Hansen Roach sixteen groups cross-section data set with a compatible weighting function and albedo data.

C 65	C 66	C 67	C 68	C 69	C 70	C 71	C 72
Pb 57	Pb 58	Pb 59	Pb 60	Pb 61	Pb 62	Pb 63	Pb 64
H_2O 49	H_2O 50	H_2O 51	H_2O 52	H_2O 53	H_2O 54	H_2O 55	H_2O 56
H_2O 41	4-T 42	3-T 43	3-T 44	3-T 45	3-T 46	4-T 47	$_2O$ 48
H_2O 33	4-T 34	4-T 35	Be 36	Be 37	4-T 38	4-T 39	H_2O 40
H_2O 25	4-T 26	Be 27	Be 28	Be 29	4-T 30	4-T 31	H_2O 32
H_2O 17	4-T 18	3-T 19	3-T 20	3-T 21	3-T 22	4-T 23	H_2O 24
H_2O 9	H_2O 10	H_2O 11	4-T 12	4-T 13	H_2O 14	H_2O 15	H_2O 16
H_2O 1	H_2O 2	H_2O 3	H_2O 4	H_2O 5	H_2O 6	H_2O 7	H_2O 8

4-T: Quadratic fuel cell 3-T: Triple fuel cell
Pb : Lead C : Graphite .
Be: Berelume.

Figure 1 . Cross-section of fuel core assembly for (IRT) reactor.

The distribution of the thermal neutron flux obtained in this work as a function of control rods radius have been represented by Fig. (2). The high depression of thermal neutron flux when the existing and the absence of the control rods from the centered of the nuclear reactor core.

2- The perturbation of Reactivity and effective multiplication factor

The variation of the rods radius will cause a big effect on the values of the reactivity and the effective multiplication factors, the relation that the reactivity can be calculated as a function of (Keff) given by the following equation :

Reactivity = (Keff-1)/Keff

Table (3) and Fig. 3 represent the variation of the effective multiplication factor and reactivity as a result of a various radius of the control rods with constant hight, For more information see references (6,7).

3- Reaction rate and the attenuation of neutrons.

The reaction rate can be calculated by using the following equation:

$$\text{Reaction rate} = \delta N \phi = \Sigma \phi$$

where: δ=represent the nuclear reaction cross-section.

N=represent the atomic number density.

ϕ=represent the neutron flux.

Σ=represent the macroscopic absorption cross-section

The neutron attenuation can be calculated by the following equation:

$$X = (-1/E) \ln \phi / \phi_0$$

where ϕ and ϕ_0 represent the attenuated and the original neutron flux respectively.

Table 4. gives the explanation results of the reaction rate and the attenuation of the neutrons.

The variation of the reaction ratye as a function of the variation to the control rod radius which represented in Fig. 4.

RESULTS AND CONCLUSIONS

1- All the results which was obtained in Table 2 indicate that the thermal neutron flux depression increase with the increasing of the radius of the control rod because the change of surface area of the matter as seen in Fig. 2.

2- The thermal neutron flux in cell (28) in the absence of Europium oxide is large and depression largely in the exist of the control rod inside cell (28) and the continuity of dectessing stay in the thermal neutron flux as a result of radius increases.

3- The neutron absorption of Europium oxide effects upon self-sustaining reaction in the nuclear reactor core and become subcritical state and can conclude that it is importance in manufactured this material can be used as a control rods for safety and guaranty of nuclear reactor.

4- The program which used in this work takes a part of the three dimension of fuel, reflector and control rod s cells, and for 16-flux energy groups that depend upon neutron transport equation solution and calculate the self-shielding effects because of the absorption of neutrons by Europium, specially in the surfaces layers and expose of internal layers of control rods to decrease the neutron flux.

5- The effective multiplication factor and reactivity decreases when we increase the radius, these results of Europium oxide masses that increases the effect upon the yield of the average nuclear fission.

6- The increasing of the rod radius should effect upon the distribution of the thermal conumn. The result should create a thermal disadvantage factor although the low of the reactivity of the thermal conductivity factor, also the thermal excitation of the material atoms had been studied in this work which increased as a result of the increasing temperature, finally, the effective resonance cross-section for the neutrons which collide with nuclei the radius of the rods must be small to avoid that damage[8].

7- The advantage of natural Europium can be saved as a gamma ray source with a high radioactivity with limit (0.4-1.4 Mev.)[9].

8- The attenuation of the neutrons will increase due to the increasing the rods radius. Therefore the reaction rate decrease as result of depression of the flux as shown in Table 4 and Fig. 4.

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Table 1 Number density of Europium oxide assembly

Radius (Cm)	Atomic density of Eu (N/Cm^3) *E-24	Atomic density of 02 (N/Cm^3)*E-24
0.1	1.5604E-5	1.2866E-2
0.2	6.2417E-5	1.2934E-2
0.3	1.4044E-4	1.3051E-2
0.4	2.4967E-4	1.3215E-2
0.5	3.9011E-4	1.3425E-2

Table 2 Thermal neutron flux as a function of control rods radius

Raduis (Cm)	0.1	0.2	0.3	0.4	0.5
Flux *E12 (N/Cm^2.Sec.)	7.387	2.044	1.131	0.4711	0.312

Table 3 Thermal neutron flux, effective multiplication factor and reactivity

Radius (Cm)	Mass (Gm)	Flux (N/Cm^2 Sec.)*E12	Reactivity	Keff.
0.1	13.520	7.387	0.03660	1.0380
0.2	54.080	2.044	0.00190	1.0012
0.3	121.68	1.131	-0.00022	0.9997
0.4	216.32	0.471	-0.00260	0.9979
0.5	338.00	0.312	-0.00502	0.9950

In the aabsence of control rods (out of the ractor core)
Thermal neutron flux = 2.1254E13 N/Cm^2 Sec.
Keff = 1.054 Reactivity = 0.0541

Table 4 The attenuation of neutron and reaction rate for several radius

Radius (Cm)	Attenuation of neutron (Cm)	Reaction rate avsor./ Cm^3 Sec.
0.1	9.52E-3	8.1996E14
0.2	2.15E-2	2.2688E14
0.3	2.68E-2	1.2550E14
0.4	3.48E-2	5.2280E13
0.5	3.85E-2	3.4620E13

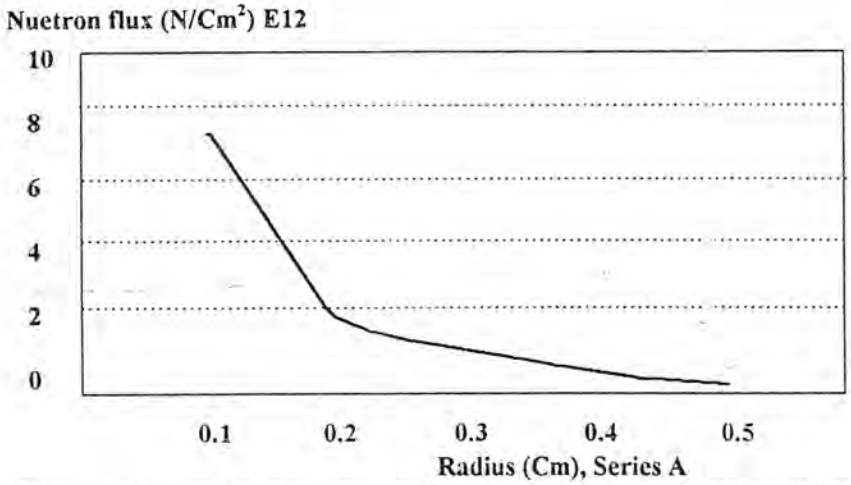

Figure 2. Variation of thermal neutron flux as a function of control rods radius .

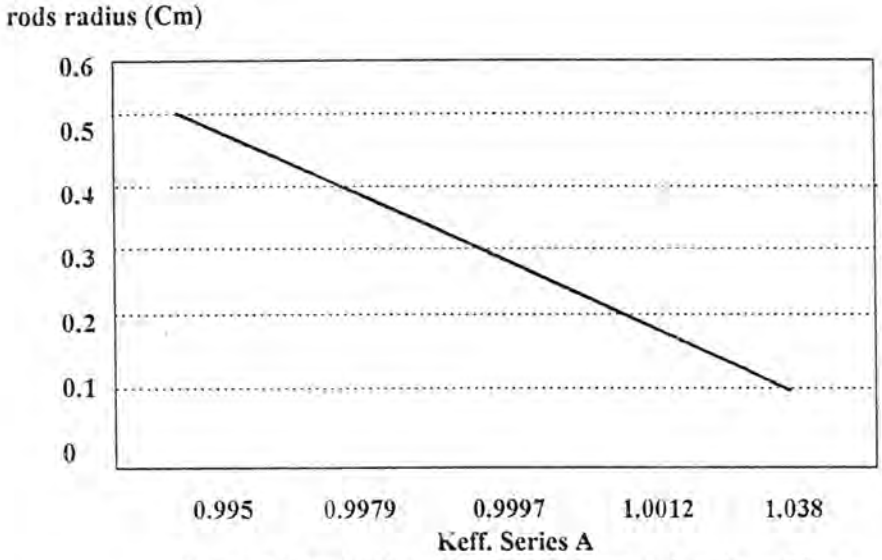

Figure 3 variation of Keff and oontri rods radius.

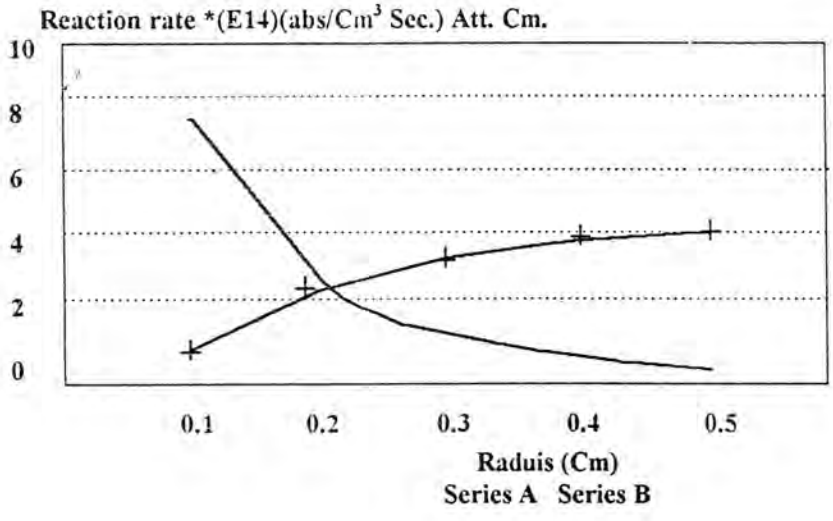

Figure 4. The reaction rate attenuation and the radius of the control rods

Nuclear Structure Study Of ^{169}Tm From The Decay ^{169}Yb

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الخلاصة

تم في هذه الدراسة قياس الطاقات والشدات النسبية للأنتقالات الكامية للنظير ^{169}Tm الناتجة بعد الأسر الألكتروني من قبل ^{169}Yb وذلك بأستخدام كاشف الجرماتيوم عالي النقاوة (HPGe) ذو الحجم 79 cm^3 لقياس طيف اشعة كاما المباشر واستخدام مطياف تطابقي سريع / بطيء يتكون من (T1) NaI - HPGe مع مميز ذو الجزء الثابت (CFD) لدراسة الأطياف التطابقية عند ست بوابات محتارة هي : 109.9 و 118.3 و 130.6 و 177.3 و 197.9 و 261.0 كيلو الكترون فولت. أكدت القياسات المباشرة وجود الأنتقال 301.5 Kev. لقد تم تحديد 27 خطاً كامياً استخدمت لبناء المخطط الأنحلالي الجديد والمتضمن على 13 مستوي طاقة . كما تم حساب عمر النصف لمستوي الطاقة 379.2 KeV باستخدام تقنية التطابق المتأخر ووجد أنه يساوي (52.5 ns). إعتماداً على قيمة عمر النصف الكلي وعلى الشدات النسبية وكذلك على معامل التحول الداخلي تم حساب اعمار الأنصاف الجزئية للأنتقالات الفرعية من هذا المستوي، قورنت النتائج التي تم الحصول عليها بتقديرات جماعياً . وقد تم حساب الأحتماليات المنحسرة لأنتقالات كامية من نوع E1 و M1 و E2 من المستويين 316.1 KeV و 379.2 . وأخيراً تم حساب عزم القصور الذاتي للنوة ^{169}Tm بالنسبة لحالتين هما كون النواة جسماً صلداً وكونها مائعاً . وتشير المقارنة بين النتائج التي تم الحصول عليها الى أن النواة لاتتصرف كجسم صلد محض ولا كمائع محض إنما كحالة وسطية تقع بين هاتين الحالتين المتطرفتين .

ABSTRACT

The energies and relative intensities of gammatransitions of ^{169}Tm after the electron capture of ^{169}Yb (31.8 days) have been measured using a 79 cm^3 HPGe coaxial detector for single spectrum and HPGe-NaI(T1) spectrometer for fast / slow coincidence measurements which are conducted at six selected energy gates : 109.9; 118.3; 177.3; 197.9 and 261.0 KeV, with the use of a Constant Fraction Discriminator technique. The singles measurements confirm the existence of the 301.5 KeV transition. A total of 27 gamma lines were identified and placed into a new level scheme consisting of 13 energy levels. The half-life of the 379.2 KeV state has been measured by the delyed coincidence method and found to be 52.5 ns. The partial half-lives of branches depopulating the 379.2 KeV level have been calculated on the basic of the total half-life, the relative intensities and the internal conversion coefficient. The results obtained compared with the weisskopf signle-particle estimates, which show a collective behaviour of ^{169}Tm nucleus. The reduced transition probabilities for E1, M1 and E2 gamma-transitions depopulating the 379.2 and 316.1 KeV levels have been calculated. Finally, the moments of inertia of ^{169}Tm nucleus have been calculated for rigid and fluid irrotational cases. A comparison between the obtained results pointed out that the nucleus do not behave like a pure rigid body nor like a fluid but in between.

INTRODUCTION

The level structure of ^{169}Tm nucleus populated in the electron capture decay of ^{169}Yb (31.8 days) has been a subject of repeated investigations by many workers (1-6) using different types of detectors. Nevertheless, some discrepancies and controversies are still exists in many gamma -transitions and in the decay schemes. In addition, earlier measurements of energies and intensities of some gamma rays are not in a good agreement with each other. Sen et al. (3) and Miminoshvill et al. (6), for example, confirmed the existance of 388; 394 and 411 KeV gamma-lines, which are not observed later in the singles spectrum given by Verma et al. (2), Mehta et al. (4). Moreover, the intensities of the 63.1; 109.8 and 177.2 KeV gamma rays detected in Ref. (5) were all higher than the values reported by other workers including this study.

For these reasons, and in order to establish a new decay scheme, we have reinvestigated the ^{169}Yb isotope employing a relatively large volume HPGe in direct measurement and HPGe-NaI(Tl) in coincidence techniques.

EXPERIMENTS AND DATA ANALYSIS

The ^{169}Yb source used in this study was obtained from the Iraqi Atomic Energy Commission (IAEC), which produced by the ^{169}Yb(n,γ) ^{169}Yb nuclear reaction. The source was a liquid in the form of yitterbium nitrate in nitric acid solution with concentration of (1mCi/1ml) and activity of (1 mCi).

The single gamma -ray spectra have been measured using HPGe coaxial detector of 79 cm^3 volume and 2.2 KeV resolution at 1332 KeV gamma-line of ^{60}Co. The detector was surrounded by 2 inch thick lead shield to avoid background radiations. The data were recorded by a 4096 MCA series 85 from Canberra possessing a three built-in microprocessors that provides several useful facilities for data analysis. The singles spectra of ^{169}Tm been measured for three times, to see the consistancy of the intensities and to check the impurity peaks, if any, and the existence of sum peaks. A typical single spectrum is shown in Fig.(1).

For accurate energy measurements, the HPGe detector was calibrated in the energy range 50-1400 KeV, using standard calibrated sources ^{152}Ba provided by the IAEA and ^{166}Ho provided by IAEC.

The gamma-gamma coincidence measurements were performed using the HPGe detector with the standard 7.6 cm × 7.6 cm NaI(Ti) detector in the fast/slow coincidence mode. The two detectors were kept at right angle to each other to reduce the crystal to crystal Compton scattering. The ^{169}Yb source was placed at 8 cm from the HPGe detector and at 12 cm from NaI(Ti) scintillation detector. The resolving time of the circuit was 9.8 ns.

The delayed coincidence technique was performed using a two 1.5 inch × 1 inch NaI(Ti) scintillation detectors. The two detectors were kept at 90° to each other with source at a distance of 5 cm between them. The Time-to-Amplitude Converter (TAC) of time analyzer was used in connection with MCA. The pulses from both scintillation detectors are amplified and analyzed for inherent energy and time information. The generated signals are fed to the start and stop inputs of the TAC. The output pulse of the TAC is digitized by ADC and stored under a selected channel number of the MCA.

The performance and reliability of all arrangement of spectrometers used in signles, fast/slow coincidence and life-time measurements are explained in details in Ref. (7).

RESULTS

3.1 Gamma-Ray Energies and Intensities

Several signles gamma-ray spectra for ^{169}Yb have been obtained for time duration ranged between (6-20) hours. Figure (1) shows a typical sugnles spectrum. In the present work, 27 gamma-tranitions have been reported, the energies and relative intensities of these transitions are listed in Table (1) and (2), together with the results of some other investigations (2,4 and 5).

The gamma transitions 81, 172, 300 and 457 KeV previously reported by Talal (5) only, are not observed in this study nor the other studies. Hence these transitions are not certain from the decay of ^{169}Yb.

The 285 and 295 KeV lines observed by Verma et al. (2) and Lederrer et al. (8), are also observed in the present study. These lines are not detected by Talal (5), but a nearby line of 287.0 KeV is detected. The 206.2 KeV gamma-ray has been detected only by Mehta et al. (4), as a transition belongs to the decay of ^{169}Yb, which is not observed in our work. On the other hand, the 207.0 KeV line detected by Sen et al. (3) is considered as a sum peak of gamma-ray 198 and LX-ray 9 KeV.

The gamma-ray 229 KeV reported by Talal (5) and Mininoshvilliet al. (6), is not detected in this study nor in other investigations. Sen et al. (3) also has not observed it, but he gives an upper limit to the relative intensity of this peak which is three times smaller than in Mininoshvili et al. (6).

The two gamma-transitions at energies of 316 and 386 KeV observed by Mehta et al. (4) are also detected in this work. Sen et al. (3) attributed the 316 KeV gamma-ray to impurities in the original source. The 379, 562 and 570 gamma-rays previously detected in ref. (4), are not observed in our measurements. The last two lines, observed by Talal (5), are embedded in the gamma-lines due to impurities of (^{152}Eu, ^{134}Cs). These impurities are not detected in our source.

The 354 KeV peak reported by Talal (5), with an upper limit relative intensity, and by Verma et al. (2), is not observed in this study. This line considered by Sen et al. (3) as a sum peak of gamma-ray of (93.7 + 261.0 KeV).

One of the latest studies on the decay of ^{169}Yb was done by J. Adam et al. (9) using an anti-Compton spectrometer has indicated several new gamma transitions with energies >300 KeV. These transitions are 500, 633, 693, 739, 760 and 781 KeV which are observed in any study including this study. We believe that a further investigation is needed solve this discrepancy.

3.2 Gamma-Gamma Coincidence Results

In order to establish a new decay scheme, conincidence measurements were performed at six selected energy gates: 109.9; 118.3; 130.6; 177.3; 197.9 and 261.0 KeV. A typical conincidence spectra are shown in Fig. (2). The expected gamma-lines including transitions in cascade are observed and those gamma lines that expected but not observed, because of their weak intensities, are all summarized in table (3). It seems to be that the 63.1 KeV and the 93.7 KeV gamma-rays are in conicidence, but weakly, with the all gates.

3.3 Half-Life of The 379.2 KEV Level

The half-life of the 379.2 KeV state of ^{169}Tm has been measured by the delayed coincidence techique. The main problem of precise measurement of the half-life of this state caused by the background of K X-ray following the conversion of gamma transitions which depopulate the 316.1 KeV state. The KX-ray and the 63.1 KeV peaks are used as

staring and stopping signals of the TAC, respectively.

It is worth to mention here that the point by point coincidence measurements with the use of different delays in the fast channel must rely essentially on the right hand side of the time spectrum where many experimental points are available and the backround contribution is distinguishable.

The half-life of the 379.2 KeV state, measured from the delayed coincidence curve shown in Fig. (3), is found to be equal to ($T_{1/2}$ = 52.5 ns). This value is in good agreement, with the measurements [$T_{1/2}$ = 52.6 ns (1987), $T_{1/2}$ = 54.1 ± 0.5 ns (1973)], which are given in Ref. (7).

The 379.2 KeV level depopulated mainly throught three branching transitions: 63.1(92%) ; 240.3 (0.13%) and 261.0 (1.8%) KeV. The fourth branch, namely, the 370.4 KeV could not be considered here in the calculations because of its weak intensity (only 0.001%). The partial half-lives of the three branches are calculated via the observed $T^{exp}_{1/2}$, the relative transition intensities I_{tot} and the internal conversion coefficients α_{tot} by the following formula:

$$T^{\gamma}_{1/2} = T^{exp}_{1/2}[1+\alpha_{tot}(a)]\frac{\sum I^{(x)}_{tot}}{I^{(a)}_{tot}} \quad \text{..... (1)}$$

The obtained results are compared with the single-particle Weisskopf estimates as shown in Table (4).

3.4 Reduced Transition Probabilities

As it is well known from the nuclear structure of ^{169}Tm nucleus that there are two isomeric states having the spin and parity $7/2^+$ (for 316.1 KeV) and $7/2^-$ (for 379.2 KeV), and that these states are linked by E1 transition having $\Delta K=0$ (63.1 KeV). The $7/2^-$ (523) states also decays by forbidden E1 transitions ($\Delta K=3$) to the $7/2^+$ and $5/2^+$ of the ($K=1/2^+$) rotational band.

In the present study we calculated the reduced probabilities of E1, M1 and E2 transitions from the 379.2 and 316.1 KeV states using a general relation given in Ref. (12).

$$B(\sigma L)\frac{C(\sigma L)}{E^{2L+1}T^{\gamma}_{1/2}(\sigma L)} \quad \text{.... (2)}$$

Where B(σL) is the reduced transition probability, C(σL) is constant for each multipolarity, E is the energy of transition in KeV and L is the multipole order. The obtained results in this study are listed in Table (5) with a comparison with the results obtained by Adam et al. (13). It is obvious that the two results are in good agreement.

3.5 Moment of Inertia Calculations

In this the nuclear structure of the odd-A ^{169}Tm nucleus has been studied by considering the moment of inertia of this nucleus in two exterme cases: the rigid and the fluid irrotational cases. The nuclear moment of inertia is obtained from the first excited state (8.4 KeV) is found to be $\rho=17.48 \times 10^{-39}$ KeV.S^2.

If the nucleus is assumed to exhibit both rigid (eq.3) and fluid irrotational motion (eq.4), the contributions of the rigid and the fluid parts of the motion for the moment of inertia depend on the nuclear deformation parameter β.

$$\rho_{rigid} = \frac{2}{5}MR^2_{av}(1+0.31\beta) \quad \text{..... (3)}$$

$$\rho_{fluid} = \frac{45}{16\pi}\rho_{rigid}\beta^2 \quad \text{..... (4)}$$

Using the value of (0.28) for the deformation parameter, the rigid and the fluid moment of inertia will be equal to ρ_{rigid} = 33.75 × 10^{-39} KeV. S^2 and ρ_{fluid} = 2.36 × 10^{-39} KeV. S^2 respectively. From these results one can assume that the

^{169}Tm nucleus do not behave like a rigid nor like a fluid body but in between.

DISCUSSION AND CONCLUSIONS

Although the results obtained in the present study are in reasonable agreement with most of the previously reported studies except of the weak gamma transitions. Nevertheless, the energies and relative intensities of gamma-rays of ^{169}Tm nucleus given by all workers indicate that discrepancies and controversies are still exists in many gamma-transitions and in the decay scheme. Therefore further investigations are still needed.

The 20.7 KeV line is not observed clearly in this study because of the effect of the Compton background. The 85.3, 291.4, 301.5 and 395.1 KeV lines are observed in the study of Ref. (5) and in this study. But we considered the first line as a background, the second line as a sum peak of (93.7 + 197.9 KeV) lines, third line as a transition between the 646.8 and the 345.0 KeV energy levels and not between the 633 and 331 KeV levels as given by Talal (5). This conclusion seems to be more reasonable because it is the only transition that feeds the 345.0 KeV level, which is also supported by Adam et al. (9), and because of its possible M1 character. The 395.1 KeV line as due to impurity in the source. The 117.3 KeV gamma-ray observed by other authors (2,3 and 4) is not oberved in the present study and in Ref. (5). This peak has weak intensity and it is probably embedded in the nearby line 118.3 KeV.

The measured half-life of the 379.2 KeV excited state id found to be equal to 52.5 ns. The partial half-lives of the three branches (63.1, 240.3 and 261.0 KeV) from the 379.2 KeV level is calculated and compared with Weisskopf estimation. The 63.1 KeV E1 transition has hindrance factor of the order of 10^5 compared with the single-particle estimate. On the other hand, the very large hindrance factor for E1 transitions of 240 KeV ($\cong 10^9$) and 261 KeV ($\cong 10^8$) could be due to high degree of K-forbidden ($\Delta K = 3$). From this comparison we can conclude the collective behaviour of ^{169}Tm nucleus.

The reduced transition probabilities of E1, M1 and E2 from the 379.2 and 316.1 KeV states are calculated and they found to be in good agreement with those given in Ref. (13). The moment of inertia of our odd-A nucleus (^{169}Tm) has been calculated, from which we can conclude that nuclei in the deformation region $150 \leq A \leq 190$ have a collective behaviour and they are not considered neither as a rigid body nor as a fluid irrotational but in between these two extreme cases.

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Table (1) Energies of Gamma-rays in (KeV) emitted in the decay of ^{169}Yb.

Eγ (KeV) Present Study	Mehta et al.(4)	Talal (5)	Verma et al. (2)
---	20.7	---	---
63.1	63.12	63.1	63.12
---	---	81.4	---
---	---	85.3	---
93.7	93.6	93.5	93.62
109.9	109.8	109.8	109.77
---	117.3	---	117.2
118.3	118.2	118.2	118.19
130.6	130.5	130.5	130.51
156.9	156.7	---	156.7
---	---	172.0	---
177.3	177.2	177.2	177.19
197.9	198.0	197.9	197.98
---	206.2	---	---
---	---	229.1	---
240.3	240.3	240.4	240.3
261.0	261.1	261.1	261.0
285.7	---	287.0	285.0
---	---	291.4	---
295.4	---	295.1	295.0
301.5	---	301.3	---
---	---	303.0	304.0
307.6	307.7	307.7	307.72
316.3	316.2	---	---
---	---	318.0	---
328.2	328.0	328.0	328.0
333.0	333.9	333.4	---
336.9	336.5	336.2	336.5
---	---	354.0	354.7
370.4	370.8	370.0	370.8
---	379.3	---	---
386.7	386.7	---	---
425.0	425.0	---	425.0
---	---	457.7	---
---	---	466.9	---
---	---	491.4	---
494.8	494.2	494.5	---
515.6	515.2	515.0	515.2
528 1	828.6	528.0	---
---	562.3	---	---
---	570.5	---	---
579 2	579.4	579.0	579.5
600.3	600.2	600.0	600.3
625 1	624.2	625.0	625.0

Table 2 : Relative intensitiess of gamma - transitions emitted in the decay of ^{169}Yb.

present study			Metha et al. [4]		Tatal [5]		Verma et al. [2]		Lederrer & Shirley [8]	
Eγ(KeV)	I %	(ΔI)	I%	(ΔI)	I%	(ΔI)	I%	(ΔI)	I%	(ΔI)
63.1	125.0	(9)	124.9	(17)	131.10	(9)	125.2	(32)	123	(5)
93.7	7.60	(8)	7.28	(10)	8.20	(2)	7.61	(18)	7.2	(3)
109.9	48.9	(3)	48.9	(5)	53.80	(3)	49.7	(9)	49	(2)
118.3	4.50	(5)	5.24	(5)	5.75	(1)	5.38	(9)	5.4	(2)
130.6	30.38	(17)	31.68	(25)	30.80	(3)	31.8	(12)	32	(2)
156.9	0.015	(7)	0.027	(1)	-		0.09	(2)	0.023	(4)
177.3	60.8	(5)	62.4	(5)	73.80	(4)	61.4	(10)	61	(3)
197.9	100.0		100.0		100.0		100.0		100.0	
240.3	0.322	(18)	0.334	(4)	0.26	(2)	0.367	(20)	0.34	(2)
261.0	4.68	(6)	4.75	(3)	5.61	(1)	5.43	(7)	4.8	(2)
285.7	0.006	(6)	-		-		0.0080	(13)	-	
295.4	0.012	(15)	-		0.115	(22)	0.020	(2)	-	
301.5	0.005	(9)	-		0.016	(10)	-		-	
307.6	27.49	(17)	27.94	(20)	33.1	(10)	30.94	(42)	28	(1)
316.3	0.003	(5)	0.009	(8)	-		-		0.0001	(3)
328.2	0.015	(7)	0.018	(12)	0.0432		0.020	(8)	-	
333.0	0.002	(3)	0.0070	(9)	0.0063		-		0.0045	(6)
336.9	0.022	(12)	0.028	(16)	0.050		0.026	(8)	≤0.02	
370.4	0.004	(4)	0.022	(16)	0.0098		0.019	(1)	0.0029	(2)
386.7	0.001	(3)	0.001	(3)	-		-		0.0010	(2)
425.0	0.002	(4)	0.004	(8)	-		0.002	(1)	-	
494.8	0.006	(10)	0.004	(8)	0.0069		-		0.0041	(2)
515.6	0.010	(7)	0.011	(8)	0.0192		0.013	(1)	0.0117	(5)
528.1	0.005	(4)	0.007	(8)	0.0033		-		0.0003	(4)
579.2	0.003	(3)	0.006	(10)	-		0.005	(2)	0.0053	(3)
600.3	0.004	(4)	0.002	(5)	0.0065		0.005	(3)	0.0032	(2)
625.1	0.008	(5)	0.011	(8)	0.0090		0.020	(5)	0.014	(1)

Table(3)results of gamma-gamma coincidence measurements in the decay of ^{169}Yb.

Gate (KeV)	Coincidence gamma-rays (KeV)
109.9	63.1[(a)]; 93.7[(a)]; 177.3; 197.9; 240.3; 261.0 (20.7; 117.3; 156.9; 285.7; 333.0; 494.8; 528.1; 515.6; 579.2; 600.3)*
118.3	63.1[(a)]; 93.7[(a)]; 177.3; 197.9; 240.3; 261.0 (20.7; 117.3; 156.9; 285.7; 295.4; 333.0; 494.8; 515.6; 528.1; 600.3)*
130.6	63.1[(a)]; 93.7[(a)]; 177.3; 240.3 (117.3; 156.9; 285.7; 295.4; 333.0; 494.8; 579.2)*
177.3	63.1[(a)]; 93.7[(a)]; 118.3; 109.9; 130.6 (20.7; 117.3; 156.9; 285.7)*
197.9	63.1[(a)]; 93.7[(a)]; 109.9; 118.3 (117.3; 156.9; 285.7)*
261.0	93.7; 109.9; 118.3

a : means weak coincidence

* : expected but not observed

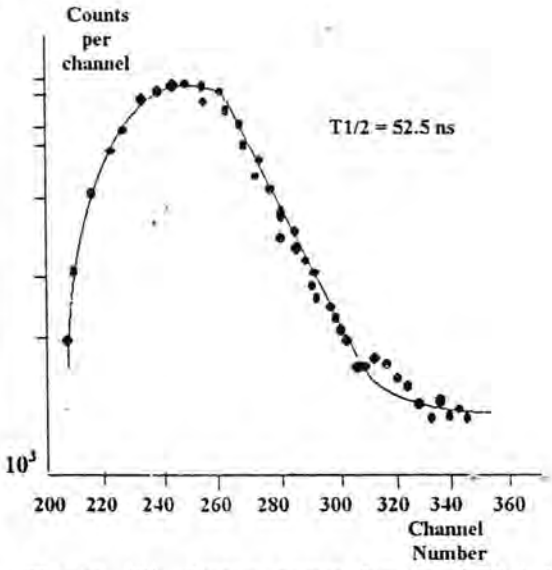

Figure (3) Decay curve for the 379.2 KeV excited excited state of Tm=169, using time-to-amplitude convertes (TAC) (^{1}ch = 2.5 ns).

Table (4) Partial half - lives of gamma - transitions depopulating the 379.2 KeV compared with the Weisskoof single particle estimate.

E (KeV)	Multipole order	$T_{1/2}^{exp}$ Sec.			$T_{1/2}^{sp}$ (Weisskopf)	$F=\left[\frac{T_{1/2}^{exp}}{T_{1/2}^{sp}}\right]$	ΔK
		Our study	Enuleseu et al (10)	Artamonova			
63.1	E1	1.14×10^{-7}	1.11×10^{-7}	0.75×10^{-7}	9.02×10^{-13}	1.25×10^{5}	0
240.3	E1	4.45×10^{-5}	3.9×10^{-5}	2.6×10^{-5}	1.63×10^{-5}	2.78×10^{9}	3
	(0.9%)M2	4.9×10^{-3}		4.0×10^{-3}	3.36×10^{-7}	1.45×10^{4}	3
261	E1	3.04×10^{-6}	2.78×10^{-6}	1.9×10^{-6}	1.27×10^{-14}	2.38×10^{8}	3
	(0.1%)M2	3.08×10^{-3}		3.0×10^{-3}	2.22×10^{-7}	1.38×10^{4}	3

Table (5) Reduced transition probabilities, depopulating the 379.2 and 316.1 KeV for E1, M1 and E2 multipoles

E_I(KeV) ($T_{1/2}$ns)	E_γ(KeV)	Order	$B(E1)^{exp}$ $e^2.b$		$B(M1)^{exp}$ $(nm)^2$		$B(E2)^{exp}$ $e^2.b^2$	
			present work	Adam et al. [13]	present work	Adam et al. [13]	present work	Adam et al. [13]
379.2 (52.9)	63.1	E1	1.53 $\times 10^{-7}$	1.50(1) $\times 10^{-7}$				
	240.3	E1+ (0.9%)M2	7.12 $\times 10^{-12}$	7.5(5) $\times 10^{-12}$				
	261.0	E1+ (0.1%)M2	8.14 $\times 10^{-11}$	8.3(5) $\times 10^{-11}$				
316.1 (660)	177.3	M1+ (15.5%)E2			2.29 $\times 10^{-6}$	2.03(23) $\times 10^{-6}$	1.7×10^{-5}	1.78(20) $\times 10^{-5}$
	197.9	M1+ (9.4%)E2			2.91 $\times 10^{-6}$	2.59(25) $\times 10^{-6}$	9.8×10^{-6}	9.0(9) $\times 10^{-6}$
	307.6	E2(Pure)					3.16×10^{-6}	3.2(4) $\times 10^{-6}$

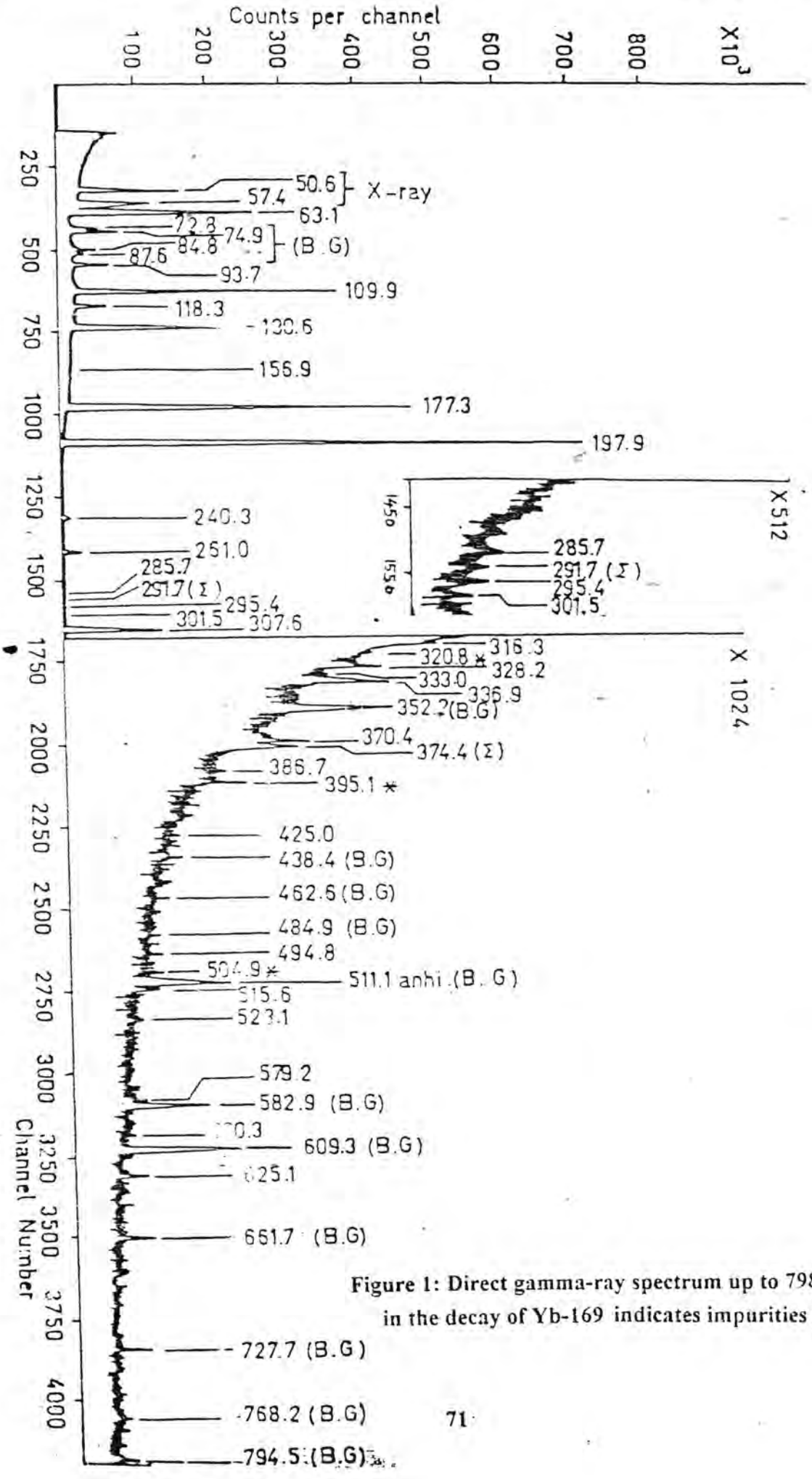

Figure 1: Direct gamma-ray spectrum up to 798 KeV in the decay of Yb-169 indicates impurities peaks.

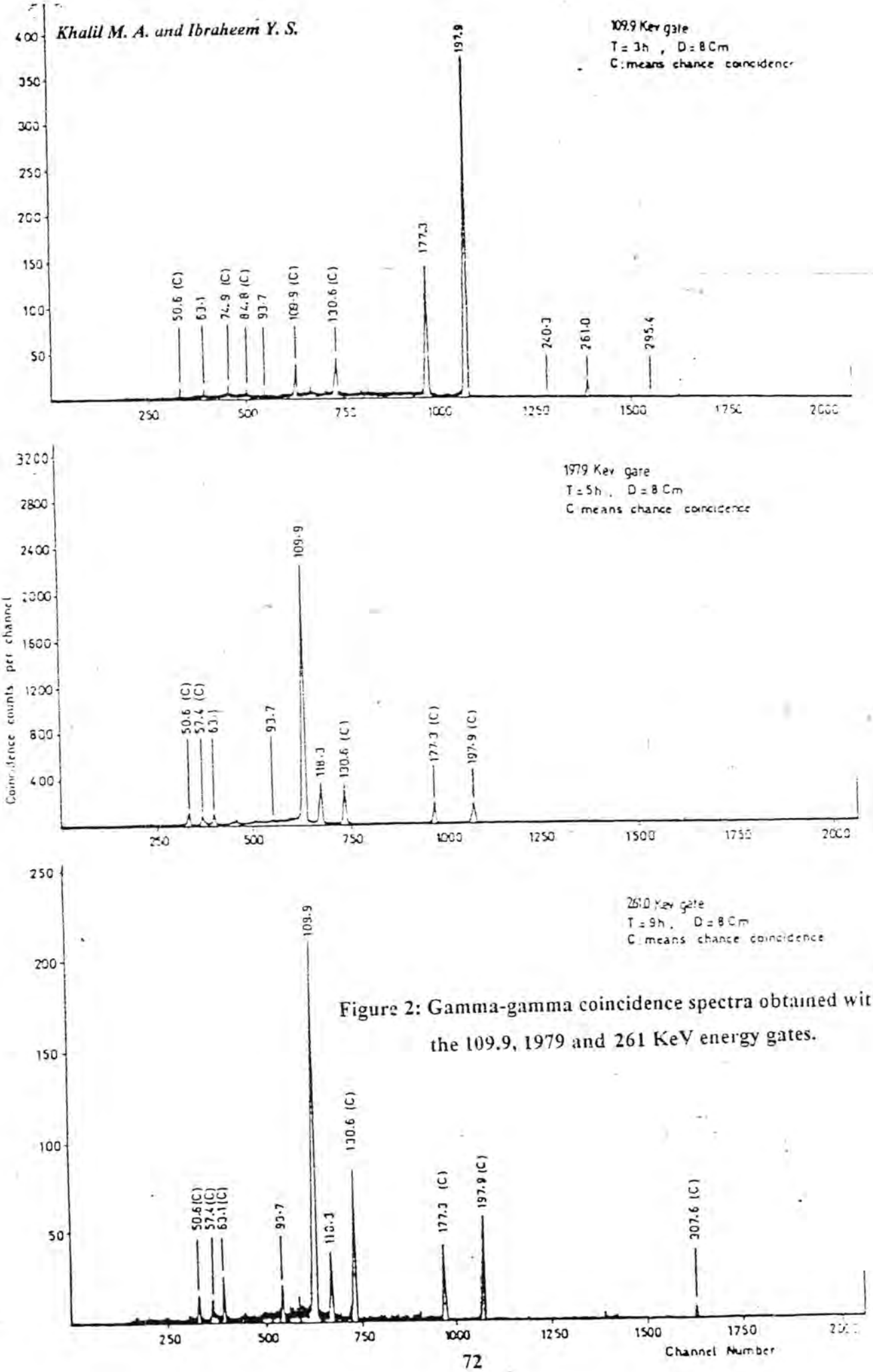

Figure 2: Gamma-gamma coincidence spectra obtained with the 109.9, 1979 and 261 KeV energy gates.

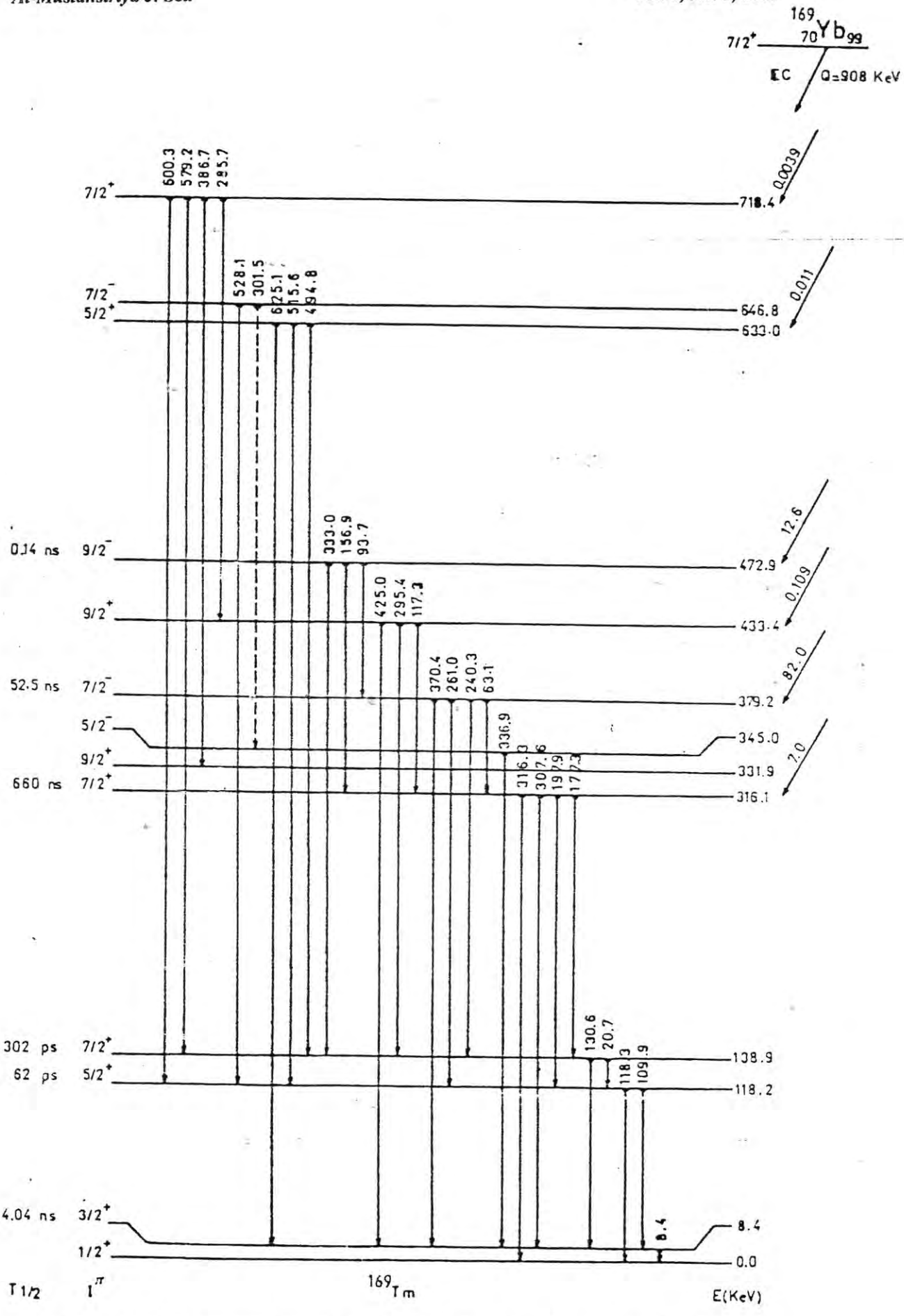

Figure 4: The decay scheme of ^{169}Tm resulting from the decay of ^{169}Yb.

The Radial Temperature Distribution in The Fuel and Control Cell Cross - Section

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الخلاصة

الامتصاص الحراري في مختلف المواد تؤخذ بنظر الأعتبار في المفاعلات النووية مثل الوقود والسموم القابلة للاحتراق زاعمدة تاسيطرة والاغطية ... الخ . الامتصاص وتوزيع درجات الحرارة مهمة في المواد المستخدمة كوقود نووي ومواد أعمدة السيطرة. هذه الدراسة تتعلق بتغيرات معدل توزيع درجات الحرارة هي الخلية وعلى طول سمك العمود . معدل الانتقال الحراري وحساباته أجريت نظرياً وبأستخدام الحاسبة . اسلوب التقريب الاتجاهي المتغير (A.D.IT) والشروط الحدودية لحسابات من هذا النوع استخدمت بهذه الدراسة . من محاسن هذه الطريقة هي دراسة تصاميم المتغيرات بصورة سريعة . قورنت النتائج مع نتائج المنحنيات من برنامج مطور يدعى MUTTR (1) .

ABSTRACT

Heat absorption in different materials are considered in nuclear reactors such as fuel, burnable poisons, control rods, clading ets. The absorption and the temperature distribution are needed for the nuclear fuel materials and for the control rod materials. The study concerned with the variations of the average temperature distribution in the cell along its thickness. Heat transfer rate calculations were caried out thertically by determind the basic programe. Alternating direction iterative teqnique and the diricklet boundary value problem are used in these calullations. The advantage of this method, even its elementary form, is its usefuless in conducting rapid design parameter studies. Results compared relatively with the results of curves obtained from a modified program called MUTTR (1).

INTRODUCTION

The problem of temperature distribution in fast breeder reactors is some what more complicated than in the case of thermal reactors.

Many of the problems of mathematical physics involves the solutions of partial differential equations. A wide variety of implicit approximations to the heat-flow equation have been studied (2).

The alternating Direction method of peaceman and ford be applied to the iterative solution of elliptic equations. The steady-state temperature in a regions containing no source of heat are studies in nuclear reactors technologeis and physics. The initial steady-state temperature satisfies Laplace's equation, which in this study is used one-dimensi-onal case so called the radial distribution of heat and the boundary temperatures given.

The work discussed the general calculation of many type geomtrical configration of the cell or fuel rods and take care with special case such as rectangular cross-section for which the

take care with special case such as rectangular cross-section for which the wall temperature and thermal conductivity are assumed known.

The programme can be used for many types of cells or chimeies and the symmetrical sides must take into account. Several approximate solutions to the heat-transfer equation for computer use. Program KER-4 and its predecessors have been used extensively in general parameter studies (3).

THEORY

The theory is based on the following assumptions and is subject to the consequent limitations.

(1)The relaxation grid size should be chosen to give two points between inside and outside surfaces.

(2)The solution will need only 1/8 of the whole crosssection since it is symmetrical.

(3)The boundary temperatures given.

(4)The thermai conductivities of cell bricks given (4).

(5)The set of simultaneous equations are written in the matrices form, such as Ax=Y.

(6)The temperature T satisfies Laplace's equations that is,

$T=0$ or $dT/dX + dT/dY\ 0$ .... (1)

We have written in rectangular co-ordinates because the plate is rectangular (5).

(7)Alternutive Direction Iterative Teqnique (1) and the dirickelet boundary value problem are used in these calculations.

CALCULATIONS AND RESULTS

(1) : Equation (1) can be cast as the form :

$S^2x * T(j,k)/hx^2 + S^2Y*T(j,k)hy^2=0$

The equations of those points in Figures (1) and (2) can be written as :

$T(j,k) = [T(j+1,k) + T(j-1,k) + T(j,k+1) + T(j,k-1)]/4$ ....(2)

$T6 = T7+T1+T1+T7/4 = 2T1+2T7/4$

$2T6 - T7 = 25$ ..... (3)

$T7 = T2+T11 + T6 + T8 / 4$

$4T7 - T6 - T11 - T8 = 25$ ..... (4)

$T8 = T7 + T9 + T3 + T12 / 4$

$4T8 - T7 - T9 - T12 = 25$ ..... (5)

$T9 = T8 + T10 + T4 + T13 / 4$

$4T9-T8 - T10 - T13 = 25$ ..... (6)

$T10 = T9 + T9 + T5 + T14 / 4$

$4T10 - 2T9 - T14 = 25$ ..... (7)

$T11 = T12 + T7 + T7 + T12 / 4$

$2T11 - T7 - T12 = 0$ ..... (8)

$T12 = T11 + T13 + T8 + T15 / 4$

$4T12 - T11 - T13 - T8 = 225$ ..... (9)

$T13 = T12 + T14 + T9 + T16 / 4$

$4T13 = T12 - T14 - T9 = 225$ ..... (10)

$T14 = T13 + T13 + T10 + T17 / 4$

$4T14 - 2T13 - T10 = 225$ ..... (11)

The results below obtaind after cast Eq.3 Eq.11 in the matrices form.

RESULTS

U(1) = 43.139631 C = T6
U(2) = 61.279421 C = T7
U(3) = 77.307443 C = T8
U(4) = 84.888424 C = T9
U(5) = 86.976454 C = T10
U(6) = 99.67067 C = T11
U(7) = 130.06985 C = T12
U(8) = 150.26985 C = T13
U(9) = 153.12902 C = T14

(2) : Cusiform temperature distribution problem :

To solve this kind can use either iteration or relaxation method and the initial condition assumed and divide the fuel rod for that shape to eight symmetricl parts and take one of them to solve by the one of the two methods which indicate above.

The fourier conduction equation can be write as :

$d^2t/dx^2 + d^2T/dY^2 + q'''/k = a\ dT/dt$ (12)

and since the oase in steady state .i.e dT/dt =0, the fourier conduction equation becomes :

d2T/dx2 + d2T/dY2 + q'''/k = 0

or

$S^2x\ Tn/hx^2 + S^2y\ Tn/hy^2 + q'''/k\ 0$ (13)

Since $Ax^2 = hx^2$ and $Ay^2 = hy^2$

Simpl\ely can be written in the finite difference Scheme (5 - point

1

1 - 4 - 1) as :

1

[T(n+Ax) + T(n-Ax) + T(n+Ay) + T(n-AY)] 4Tn + q'''Ax^2/k = 0

Where the initial conditions are :

q'''(n) = volmetric heat source.
k = Tharmal conduction factor.

For any (q'''(n) or (k) the set of equations cam be solved with initial guess mesh temperature by iteration or relaxation method, for examole if the initial condtion are :

q''' = 10E7 BTU / hr. ft^3.
k = 19.84 BTU / hr. ft. F.
Ts = 600 F ; Ax = 0.2 "
Tg = 140 F

The woeking formula after return to Figure (4) simpely becomes

Tn=([T(n+Ax) + T(n-Ax) + T(n+Ay) + T(n-Ay))]/4) + 35 ..... (14)

Then the set of equations can obtained as below :

T1 - T2 = 35
(1/4)T1 - T2 + (1/4)T3 = -335
(1/4)T2 - T3 + (1/4)T4 = -335
(1/4)T3 - T4 + (1/4)T5 = -335
(1/4)T4 - T5 + (1/4)T6 = -335
(1/4)T5 - T6 = -485

After inter the set of equations above, the results are :

T1 = 717.687 F.
T2 = 682.687 F.
T3 = 673.063 F.
T4 = 669.570 F.
T5 = 665.218 F.
T6 = 651.304 F.

Table 1 : Relative comparsion values between the results obtained and the results from references (1) for arbitrary points.

	Temp. C	Temp. C from MUTTR curves
(1) Unit cell :	T6=43.13	45
	T9=84.88	88
	T11=99.67	106
	T12=130.06	135
	T13=150.26	157
(2) Cusiform unit cell :	T1=717.68	725
	T2=682.68	685
	T3=673.06	680
	T5=661.21	669

CONCLUSIONS

Conclusions reached in this study attempted to provide a unified basis for the study of Alternative Direction Iteration Techniqe. The results indicate that temperature distribution every point of Figure (2).

The rate change of the temperature with distance as it move away from the starting point. The finite difference scheme (5-points) used in this techniqe and the difference equations is applied at the mesh points.

The gradient of a function has useful geometrical and physical meanings which investigate (6).

By used the mesh points and the number of radial zones-division. The programme calcul -ates the temperature across each pointe.

Figure (4) shows two curves of gradiente of temperature from inner side to outside. The lower curve shows the temperature gradiante from inner corner to the corner.

Devlopement of this study can be satisfied to calculations multishell temperature distributions for anuular or solid cylindrical fuel rods and control rods with either constant source calculations or with any flux depression model for grayer (more heavily loaded) fuels (1).

The results obtained compared with the results of MUTTer as listed in table (1).

The program MUTTR used a numerical-integration in order to yield proper information about temperature distribution, therefore, the results differ from that obtained from Alternating direction iterative teqnique by more than 5%. It noted that the results obtained using A.D.I.T. can not be compared directly with the peak temperature obtained by Hansen because of the different values of the thermal conducivities of the two studies (7).

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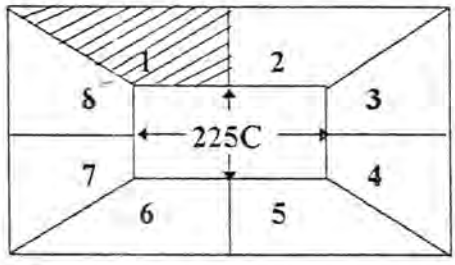

Figure. 1 : Whole cross - section of unit cell.

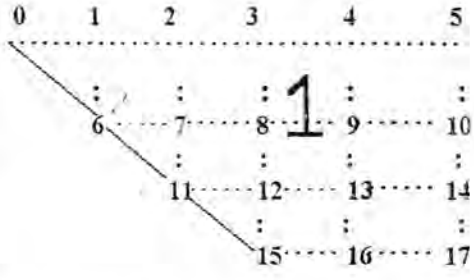

Figure. 2 : 1/8 of the whole cross-section of the symmetrical grid.

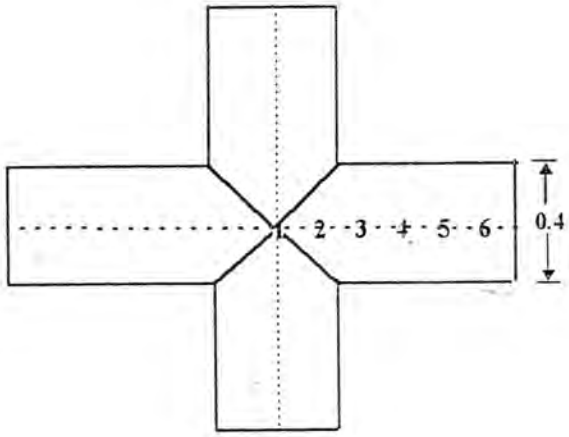

Figure. 3 : Whole cross-section of Cusiform unit cell.

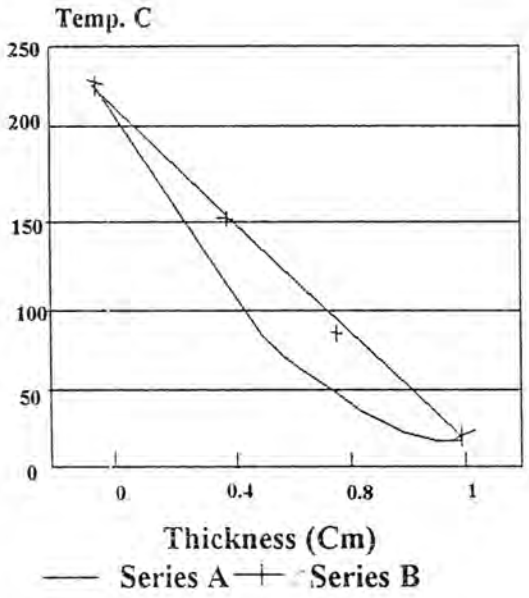

Figure 4 : Temperature distribution as a function of distance.

Photochemical Reactions of 9,10 - Phenanthrene Epoxide

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الخلاصة

تم تحضير عدد من مشتقات الفيناثرين الجديدة بواسطة التفاعل الكيميائي الضوئي لايموكسي الفيناثرين . تعتمد المواد الناتجة على نوع الآصرة التي تكسر في حلقة الايبوكسي . والتي تعتمد بدورها على كل من المذيب والمجاميع المعوضة الغنية بالكترونات وكذلك نوع المحفز الذي يستخدم بالتفاعلات الكيميائية الضوئية . ان دراسة التحليلات الطيفية للنواتج أدت الى معرفة ما هية المركبات الوسطية المتوقعة وميكانيكية التفاعلات الضوئية .

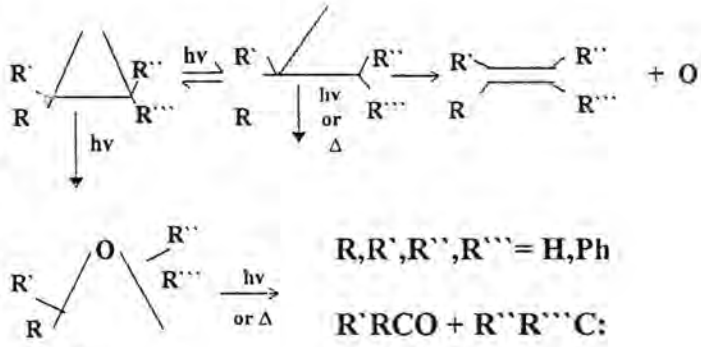
ABSTRACT

A number of new derivatives of phenanthrene compounds were prepared by the photochemical reactions of 9,10 - phenanthrene epoxide which gave different products proved to be dependent on solvent, the electron rich substituentsand the sensitizer used in the photochemical reactions. The study of the speourosepic analysis of the products enabled the determination of a number of intermediates in these systems.

INTRODUCTION

The chemistry of epoxide ring recervied a great attention during the last twenty years through their ring opening reactions[1,2]. Epoxied owe tireir selectivity to the highly strained nature of three membered ring.

The (C-C) bond seiddion of the epoxied is the most artractive mechanism for the three membered heterocyclic compounds. The excited state studies and the nature of the intermediates forms such as biradicals pairs, and ion pairs are currently of great interest in this field[1,3]. The proposed reaction paths for the epoxied are based on processe.



Scheme (1)

described for the analogous compounds, such as cxirans[1] (Scheme 1). In a variety of techniques such as spintrapping and spectroscopy analyses were demonstrated to give a new proof to this kind of photochemical reactions mechanism. This mechanism based on biradicals or ion pairs which forms the product (or products) and this dependent on the life

products) and this dependent on the life time of these intermediates. In the present work, a series of hotochemical reactions were caried out in different olvents and sensitizers in order to study the effect of substituent on the photochemical ring opening reaction of the epoxide ring.

EXPERIMENTAL

Infra-red spectra were recorded on Pye Unicam(SP-100) spectrophotomater. Melting points were determined with electro-thermal melting point apparatus and are uncorrected. Reactions were monitored by Thin-Layer Chromatography. Nereck Kiessel Gel 60 O.F 254.column chromatography was used with Fluka kiessel gel 60 as stationary phase and suitable solvents.

Medium pressure nercury lamp (250), and wave length (313, 335, 365) was used to irradiate the reaction mixtures in a pyrex vessel reaction. All the reaction mixtures were carried out under N_2 gas, and room temperature.

9, 10-Dibromo - phenanthrene :

A solution of phenanthrene (B.D.H Chemicals) (1.78 gm) in cyclohexane (10 ml) was mixed with bromine (1 ml) and the mixture was stirred at room temperature for 3 nrs. The product was obtained by recrystallization from cyclonexane (1.9 gm, 87%), m.p 87-88 °C (lit. [6] 87 °C .

9-Bromo, 10-methoxy pnenanthrene:

Bromo methoxy phenantn -rene was prepared adding 9, 10-dibromo phenanthrene (1.7 gm) (in mathylene chloride (10 ml)) to sodium mothoxide (sodiun 0.5 gm in methanol) dropwise over 30 min period. The mixture was reflexed for anather 1 hr. The product was obtained by recrystallization from cyclohexane as white crystals (0.55 gm; 35%)m. p, 79-81 °C (lit.[6] 80-81 °C).

Epoxidation of 9-Bromo, 10-Menthoxy phenantzene:

Phenanthrene epoxide was prepared by reflex of 9-Bromo, 10-methoxy phenanthrene (1.45 gm) in methanol (20 ml) in presence of sodium aydroxide (0.2 gm) for 6 hr. 9, 10-epoxy phenanthrene was recrystallized from cyclohexane as white crystals (0.48 gn; 41%) m.p 51-53 °C. Vmax. (film) 3020, 1610, 1510, 1300, 1290, 1200, 1020, 860, 810, 670 cm^{-1} . (Found: C, 97.90% H, 4.5%, $C_{14}H_{10}O$ requiras: C, 86.57%; H, 5.19% .

Epoxidation of 9, 10-phenanthrene:

Phenanthrene epoxide was prepared directly by reaction of 9, 10-phenanthrene (0.9 gm) with dobenzoyi peroxide (1.1 gm) in methylene chloride (20 ml) under dry conditions. 9, 10-epoxy phenanthrene was collected using silica dry column. with 2:1 cyclohexane and mothylene chloride as solvent. The product is white crystals (0.22 gm; 20%), m.p. 51-53 °C .

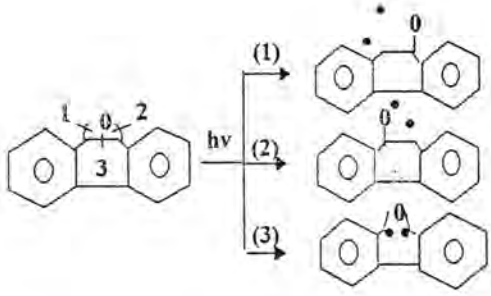
RESULTS AND DISCUSSION

The epoxidation of 9, 10-phenanthrene was carried out by two ways, First, through cyclization of halohydrins in presence of a base and second routs through peroxidation of carbon - carbon double bonds by paroxide. The first route gave higher yield (24%), even through it is three step reaction.

The photochemical race on of small intact molecules from large organic molecules has inharent interost for a variety of reasons. There are important qyestions about the nature of the exited

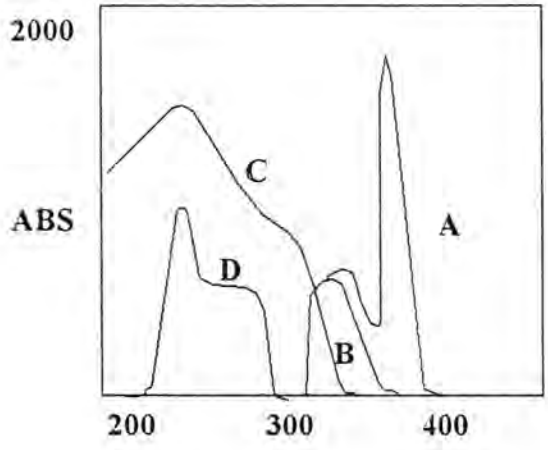
state, the energy utilization in the bond breaking and the nature of the intermejiates.

There are sevsral possible mechanisms[5] for the ring opening of epoxide by the action of the light, and these depond on the sensitizer energy and the solvent used in the reaction (Scheme-2).



Scheme-(2)

The most frwquently proposed structure for the C-e bond soission is a biradicals. The formation of the alcohol products due to C-O bond seission involving either zwitterions cr biradical intermediatcs, the yield was increased sharply by the sensitizers. When acetonc, Bromo-trifloroetherat or acetophenone used as a sonsitizer, the stability of the intermediates is incxeased (Scheme -3).



The alcohols product which was produced by photochemical reactions seams to be affected by the polarity of the solvents (Table 1). The time of intermediates which have analogous strudture icrcases in going from cyclohexane to bonsene the protic solvents (ethanol and t-butyl alcohol). The nature of that system leads us to expect that the 9, 10-phenanthrene epoxide intermediate might be affected by solvent polarity.

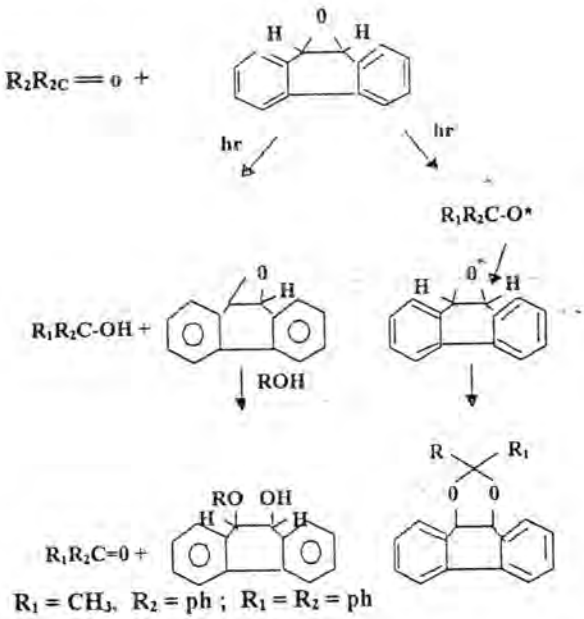
The life timc of the transient formed by photolysis of phenanth -rene epoxide increased with increasing solvent polarity. However, the properties and chemistry as well as machanistic pathways leading to this biradical are not well understood.

Thephotochemical reactions of 9, 10-phenanthrene epoxide with different alcohols (Table 1) afforded different prodncts from the C_9-O and C_{10}-O bonds rupture. The less polarity alcohole gave less yield. No further identifications were carried out to specify which bond was to break (Scheme-2).

The photochemical of the phenanthrene epoxide in asetone solution was also studied. Acetone acts as solvest triplet sensitizer. Irradiation of epoxide under these conditiel afforded two products : 2,2-Dimethyl, 1,3-diexo 4,4-phenanthrene derivative from the C-O bond seission and an unidentified product which was yieled from the fragmentation of unstable C-C bond rapture products. Nc further identification was carried out because of the unstability of the products.

The high yield of the unidentified product seems to be due to the C-C carbon rupturs as the diamer of the phenanthrene which was increased by increasing the time (Table 2).

Table 1, 2 shows that no product was detected without adding one of the sensitizers which is lead us to pridict that no photochemical reaction was occur as aresults of direct light absorption by Phenanthrene it self (Scheme-4).



Scheme-4 Wechahistic pathways of photochemical reaetions of phenanthrene epoxide

The activity of the phtochemical reaction of the carbonyl compounds (is a sensotizers) might be depend on several factores; nature of triplet excited state (T_1), nolecular structure, dissociation energy of the bond, and nature of the solvent. Triplet state (T_1) of (n-π^*) transfer leads to an active photoreduction

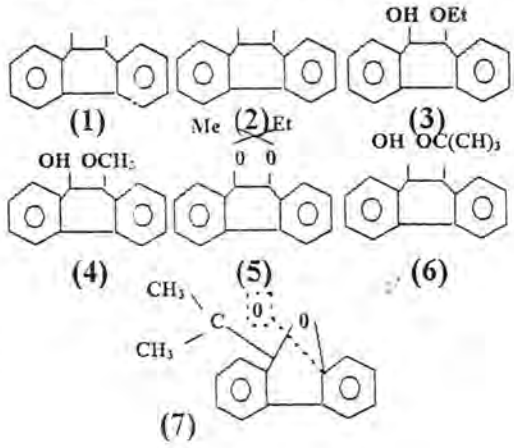
π

π^*) transfer leads to an species processes. The (n-π^*) transfer of carbonyl group will give an electrophilic atom and the(π π^*) transfer will give a nucleophilic.

The photochemical reactions of 9, 10-phenanthrene gave products only from cleavage of C-O bond of epoxide ring. No evidence was obtained for the formation of a product from C-C bond cleavage pathway. The phenyl group might be resposible for this mechanism since it provides greater stabilization of the ionic intermediates from C-O bond repture.

The high yield of the unidentified product perhaps suggests to us that the / less stabilized intermediate underges self-fragment ion due to its unstability as was the case in the C-C bond cleavage intermediates.

The above reactions were repeated under the same conditions using thermal readtions by refluxing the mixture for six hours. A traco products of alcohols were detected with the photoreaction gave a high selective yield (Table 3).



Scheme (5) Chemical strusturst of the products 1 to 7

CONCLUSION

9, 10- Epoxy Phenanthrene was prepared and irradiated in the presence of a number of different catalyst and solvents. It was expected that the alcoholic solvents give products of an alcohols as a result of C-O bond seission, on the other hand reactions with ketones give products as a result of C-C bond seission. The alcohol give the expected results, but the ketones did not. The product of ketones was a products of C-O bond seission too. The high percentage of unidentified products (which were difficult to separate) were due to C-C bond seission. The photochemical reactions of Epoxy phenanthrene needs further investigations to improve the conditions, and study the mechanisim.

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Table 1 : I.R spectroscopy and C.H analysis of the products.

Product No.	Formula	m.p. °C	I.R ν_{max} (cm^{-1})	C.H. analysis
(2)	$C_{17}H_{16}O_2$	72.74	2980 (s), 1590, 1450,1390, (ArH),1140, 1080 (C-O)	Found:C,79.0 ; H, 6.9 Requires C, 80.9; H, 6.3%
(3)	$C_{16}H_{16}O_2$	109.111	3420-3400 (O-H), 2960, 1210,1190,1120, 1020, 900, 84	Found:C, 81.5; H, 7.6 Requires C, 79.9; H, 6.7%
(4)	$C_{15}H_{14}O_2$	115.117	3440-3420, 2980,1590,1440 ,1120,1020, 920	Found:C, 75.2; H, 5.3 Requires C, 79.6; H, 6.2%
(5)	$C_{18}H_{18}O_2$	103.105	2990-2975, 1580,1480, 1450, 1140,1080,1020, 720	
(6)	$C_{18}H_{20}O_2$	109.114	3010-2980, 1410,1330,131 20,920,720	

Table 2 : Photolysis of 9, 10-Epoxyphenanthrene with different solvents under N_2

Reaction	Solvent	sensitizer	Time (hr) of irradition	Products			Unident
				1	2	5	
1	acetone	-	1	42	trace	-	41
2	acetone	A	1	29	9	-	62
3	acetone	A	2	17	8	-	75
4	acetone	B	4	-	47	-	53
5	methylene chloride	-	2	59	-	-	41
6	methylene chloride	C	4	49	8	-	43
7	methylene chloride	A	4	29	-	-	61
8	methylene chloride	B	4	27	-	-	63
9	ethyl methyl ketone	-	2	57	-	trace	57
10	ethyl methyl ketone	A	4	trace	-	21	72
11	ethyl methyl ketone	B	4	trace	-	41	56

A:Acetophenone, B:Boron trifluoroetharate, C:Acetone.

Table 3 : Photolysis of 9, 10-Epoxyphenanthrene with alcohol solvents under N_2

Reaction	Ethanol	Sensitizer	Time(hr) of irradtion	Products %				Unident
				1	3	4	6	
1	ethanol	-	2	41	21	-	-	38
2	ethanol	A	4	16	23	-	-	61
3	ethanol	B	4	-	71	-	-	29
4	methanol	-	2	51	-	17	-	32
5	methanol	A	4	19	-	26	-	55
6	methylene	B	4	-	-	57	-	43
7	t-Butanol	-	2	57	-	-	trace	40
8	t-Buta	A	4	42	-	-	9	45
9	t-Butanol	B	4	37	-	-	11	50
10	t-Butanol	B	6	trace	-	-	12	87

A:Acetophenone, B:Boron trifluoroetharate, C:Acetone.

Table 4 : Thermochemical reaction of 9, 10-Epoxyphenanthrene.

Reaction	Solvent	Sensitizer	Time (hr) of irradition	Products			Unident
				1	2	3	
1	acetone	-	2	19	trace	-	80
2	acetone	BF_3/etharate	2	7	8	-	85
3	acetone	BF_3/etharate	4	-	9	-	95
4	ethanol	BF_3/etharate	2	5	15	-	78
5	ethanol	-	2	17	trace	-	85
6	ethanol	BF_3/etharate	4	-	9	-	90

Carbon-13 NMR of Some Pharmacoloqically Important N-(4-amino-2-butynyl) Succinimides.

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الخلاصة

تهتم دراسة وتحليل اطياف الرنين النووي المغناطيسي للكاربون -١٣ لمركبات N-بروباكيل سكسنماید ذات الاهمية الدوائية ومن تلك الدراسة امكن ايجاد تاثير استبدال البروتون الاستيليني بمجموعة N-مثيلين امين حلقي في اطياف تلك المركبات

ABSTRACT

Carbon-13 NMR spectra of N-pro argyl succinimide, nine of N-(4-amino-2-butynyl)succinimide citrate were analyzed. The effect of replacing the acetylenic proton in N-Propargyl succinimide by N-methylene substituted cyclic amines is discussed.

INTRODUCTION

A number of compounds containing both amino and ethynyl functions have been reported to posses a potential pharmacological value[1]. During our investigations of acetylenic amine N-oxides the NMR spectra of some acetylenic tertiary amines and their N-oxides, N-alkynyl cyclic amines(3) N-propargyl phthalimides (4) were reported. In this work we report the carbon-13 NMR analysis of some N-(4-amino-2-butynyl) succinimides which have been found to be potent in blocking the motor effect of the muscarinic agent oxotremorine(5).

EXPERIMENTAL

N-propargyl succinimide[1] was prepared according to the previously reported method from the reaction of propargyl bromide with sodium succinate[1].

N-(4-amino-2-butnyl) succinimides 2(a-j) were prepared by Mannich reaction starting from N-propargyl succinimide, paraformaldehyde and the appropriate cyclic amines [2].

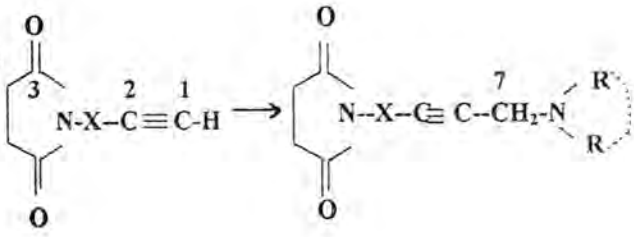
Oxidation of the acetylinic amines with m-chloroperbenzoic acid produced the corresponding tertiary N-oxides[6].

The samples were prepared in 10 mm O.D. Wilmad NMR tubes in deuteriochloroform. 2-Citrate was dissolved in DMSO-d6 (15-25% w/w).

A small amount of TMS was added to the samples as an internal standard whereas $CDCl_3$ serves as an internal deuterium lock signal source.

The carbon-13 NMR spectra were recorded at ambient prob. temperature on a Bruker WH90 DS spectrometer operating at M22. 63 MHz. The broad-

operating at M22. 63 MHz. The broad-band proton decoupled 13C NMR spectra were run at a spectral width of 6KHz & data memory size of 8K with pulse width of 6 us., line braiding of 0.6 and pulse delay of 3s. Gated decoupled spectra were used for signal identification.



RESULTS AND DISCUSSION

ASSIGNMENT OF C-13 CHEMICAL SHIFTS:

Some general methods have been used for the assignment in which protonated and non-protonated carbons are easily distinguished by different realization times and comparison throughout the series and also by the previously reported data for analogous compounds[4,5].

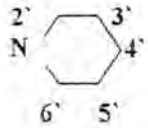
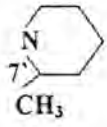
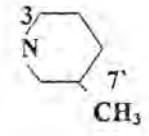
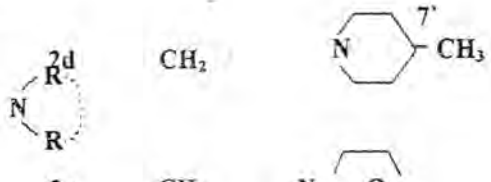
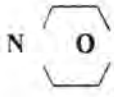
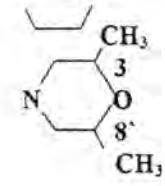
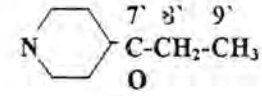
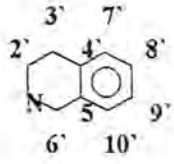
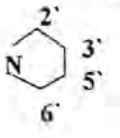
In N-propargyl succinimindes[1], the succinimide part showed two singlets at S 176.3 ppm and 28.3 ppm which were assigned to the carbonyl carbon (C-3) and to the CH_2 (triple triplet)(JCH=(135)) and ^3J=5.2 Hz respectively as shown in figure (1).

The second doublet of triplet at δ 27.6 (^2J=145.2, J=4.4 Hz) was assigned to the carbon (C-5), whereas the signal at δ 77.1 (double triplet ^1J=253 and ^3J=4.4 Hz was assigned to the protonated acetylenic carbon (C-1). Moreover the signal at δ77.5 ppm was assigned to the quaternary carbon (C-2).

Replacing H in compound (1) by methylene cyclic amine resulted in compound 2(a-j).

The spectrum of the N-propargyl succinimide moiety in compound (2) was assigned by analogy to compound (1), whereas that for the N-methylene cyclic amine portion was assigned by analogy to the same part in N-propargyl phthalimide that was previously reported[5] (Figure 2).

(1) (2)

Compound	X	
2a	CH_2	
2b	CH_2	
2c	CH_2	
2d	CH_2	
2e	CH_2	
2f	CH_2	
2g	CH_2	
2h	CH_2	
2j	CH_2-CH_2	

The assignment of (C-1) (both are quaternary acetylenic carbon) was confirmed by the variation in their chemical shifts. (C-1) had no or only very small changes through out the series while (C-2) showed a larger effect (Table).

During preparation of compound (2f), the 3,5-dimethyl morphline used as a cyclic amine was a mixture of cis and a trans isomers. Accordingly the carbon-13 NMR spectrum of compound (2f) clearly showed both isomers with the trans form being more predominate (76%). The major signals for the trans isomers are indicated in the table (the uper row), where the dis isomer (24%) showed different signals (the lower row in the table). Only the N-methylene-3,5-dimethylmoropholine carbons showed two sets of signals (i.e. the cis/trans effect was not transmitted to the succinimide portion of compound (2f).

The carbon-13 NMR spectrum of N-(5-pyrrolidino-3-pentynyl) succinimide citrate (2j) were analyzed by analogy to the other member of the series (2a-i). The citrate portion of (2j) showed two signals for the carbon of the carboxyl group at δ 176.8 ppm (COO carboxylate anion) and δ 171.8 ppm (2X COOH). The CH_2 and the CHOH carbons gave signals at δ 42.4 ppm and δ 84.7 ppm respectively. It is worth-noting that the conversion of compound 1 to compound 2 resulted in deshielding of the signals for C-1 and C-2 by 1.6+0.3 ppm and 0.5+0.6 ppm respectively, whereas the succinimide carbonyl and the CH_2[6] s (C-3 and C-4) suffered no or very small effect.

Oxidation of the acetylenic tertiary amines 2(a-j) with m-chloroperbenzoic acid produced the corresponding N-oxides which were unstable compounds and thus their C-13 NMR spectra could not be obtained.

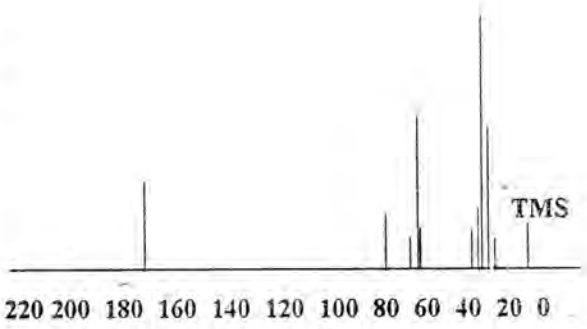


Figure 1. C13-NMR of N-Propargyl Duccinimide.

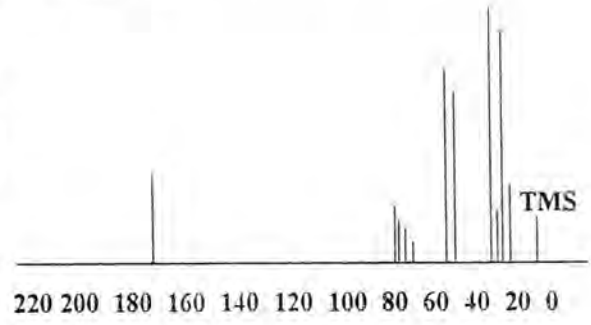


Figure 2. C13-NMR of N(-4-Pipredino-2-butynyl) Succinimide (2a).

Table 2. Carbon-13 NMR of N-(4-amino-but-2-ynyl) succinimides.

	C-1	C-2	C-3	C-4	C-5	C-6	C-7	C-2`	C-3`	C-4`	C-5`	C-6`	C-7`	C-8`	C-9`	C-10`
	77.1	77.5	176.3	28.3	27.6	-	-	-	-	-	-	-	-	-	-	-
a	78.8	77.8	175.8	28.3	28.1	-	47.9	53.5	25.9	24.0	25.9	53.5	-	-	-	-
b	78.2	78.0	175.7	28.3	28.1	-	43.5	54.9	34.8	24.4	26.4	53.3	19.8	-	-	-
c	78.9	77.8	175.7	28.3	28.1	-	47.7	60.9	31.02	32.6	25.3	52.9	19.6	-	-	-
d	78.7	77.7	175.8	28.3	28.0	-	47.4	52.8	34.3	30.3	34.3	52.8	21.8	-	-	-
e	78.6	78.2	175.9	28.3	28.1	-	47.4	52.4	66.9	-	66.9	52.4	-	-	-	-
f*	78.3	78.0	175.8	28.3	28.0	-	47.0 47.4	58.2 57.4	58.2 57.4	-	58.2	71.6 66.4	19.2 18.2	-	-	-
g	78.3	78.1	175.4	28.3	27.7	-	47.4	28.2	28.2	60.2	28.2	51.7	174.4	40.7	144	-
h	78.6	77.6	175.9	28.2	27.9	-	47.0	51.9	51.9	-	-	51.6	168.3	21.3	-	-
i	78.1	78.1	175.8	28.1	27.9	-	46.9	24.1	24.1	133.8	134.5	54.5	128.6	126.5	126.1	125.1
j**	71.9	72.8	177.6	28.0	36.4	16.9	43.9	23.6	23.6	-	23.6	52.1	-	-	-	-

*mixture 100% cis & trans isomers

** BL 14 citrate

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Excess Molar Volumes And Excess Visccosities of Binary Mixtures of n-Butanol + Isomers of Butanol at 303.15K.

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الخلاصة

لقد تم دراسة الخواص الثرموديناميكية لمحاليل ثنائية من الكحول البيوتيلي والايزومرات لهذا الكحول وهي الكحول الايزوبيوتيلي والكحول النيرشلي بيوتانول والكحول ثنائي بيوتانول وهذه الخواص تتضمن الحجوم الفائضة V^E واللزوجة الفائضة η^E في درجة حرارة ٣٠ م° وقد وجد بأن نتيجة للتجاذبات بين جزيئات هذه الكحولات والناتجة من الآصرة الهيدروجينية تعطي قيم سالبة للحجوم الفائضة واللزوجة الفائضة .

ABSTRACT

Densiti s, viscosities for the binary mixtures of n-butanol+iso, sec-, and tert-butanol were measured at 303.15k, and then excess molar volumes V^E, Excess molar viscosities η^E were calculated over the whole mole fraction range. The mixture exhibit negative V^E and η^E which suggest the molecular interaction between n-butanol and other isomers of butanol from a sort of hydrogen bonding system, this behavior was discussed on the bases of shape and position of Hydroxylic group of isomers of butanol.

INTRODUCTION

Excess molar quanities has extensive application in studies, of molecular interaction in Binary mixtures including those leading to Hydrogen bonds system form -ation[1-5] . Butanol isomers were used recently in automotive fuel technology as oxygenated products[6]. The use of alcohol's as oxygenated fuel based on Binary mixtures with other gasoline fuel minimize the environmental pollution. The present paper reports the measurements of desities, viscosities, excess molar volumes excess molar viscosities of binary mixtures of n-butanol+iso-, sec-, and tert-butanol at 303.15K.

EXPERIMENTAL

Materials :

n-butanol, iso-butanol, sec-butanol, and tert-butanol (Fluka. A. G. Pure grade) were used without purification. Purity of the Alcohol's was confirmed by the density and viscosity measurements at 298.15K and found to be in good agreement with the values published in literature[7] (Table 1).

Preparation of mixture :

Alcohol mixtures were prepa -red by weigthing out the two isomers using a 4 digits balance to yield the exact mole fraction.

Viscosities were measured using a suspended Ubbelohde viscometer (MGW Lauda, W. Germeng). The bath temperature was controlled ± 0.01K (Jobo, Type 3630 GB Germany). The flow times were determined electronically using

an electronic timer (MGW Lauda, W. Germany). with a precision of ± 0.01 sec. (the flow time of pure alcohols are more than 200 sec).

Measurements were repeated until three consecutive reading differ only by ± 0.02 sec. were obtained.

Density measurements :

Densities of the pure liquids and binary mixture of the present systems were measured at 303.15K with the calibrated Density bottle[8] (25.284ml), temperature was maintained at 30C° ±0.01C°. The accuracy in the density values measured using the density bottle is estimated to be better than 3×10^{-4} gcm^{-3} .

Table 1 : Densities ρ and Viscosities η of pure isomeric Butanol at 298.15K.

Component	ρ (observed) (g cm^{-3})	ρ (literature)[7] (g cm^{-3})	η (observed) (cp)	η (literature)[7] (cp)
n-Butanol	0.80667	0.80600	2.381	2.379
Iso-Butanol	0.79789	0.79782	2.957	2.919
Sec-Butanol	0.80214	0.80260	2.771	2.700
Tert-Butanol	.078089	0.78112	4.023	4.011

RESULTES AND DISCUSSION

The experimental values of excess molar volume V^E results obtained from precise density measurements for the systems n-butanol + tert-butanol, n + butanol + iso - butanol and n - butanol + sec - butanol at 303.15K are given in Tables 2,3 and 4 respectiviely and plotted in Figure (1).

The values of these binary mixture were calculated using the following equation[9] .

$$V^E\left(cm^3 mole^{-1}\right) = \left(\frac{M_1X_1}{\rho_m} - \frac{M_1X_1}{\rho_1}\right) + \left(\frac{M_1X_1}{\rho_m} - \frac{M_2X_2}{\rho_2}\right)$$

where X_1 and X_2 are the mole fraction of n-Butanol and butanol isomer . M_1 and M_2 are the molecular weights of n-Butanol and butanol isomer respectively, ρm is the density of the binary mixture.

The experimental values of excess molar vicosity η^E at 303.15K for these system are also given in tables 2,3 and 4. The values of η^E, of these binary mixtures were calculated using the following equation (4, 10-11).

$$\eta^E(cp) = \eta_m - X_1\eta_1 - X_2\eta_2$$

where η_1 η_2 and η_m are the viscosity of n-Butanol, isomer of butanol and of the binary mixture repectively. The results of η_m are plotted in figure (2).

Table 2 : Densities, vicosites, excess molar volumes V^E, and excess viscosites η^E for mixtures of (x) tert-butanol + (1-x) n-butanol at 303.15K.

X	ρ ($g\ cm^{-3}$)	V^E ($cm^3\ mole^{-1}$)	η (cp)	η^E(cp)
0.0000	0.80153	0.0000	2.30000	0.000
0.1509	0.79869	-0.1653	2.42206	-0.046
0.3000	0.79516	-0.2415	2.58412	-0.052
0.4501	0.79169	-0.3250	2.75918	-0.045
0.5501	0.78923	-0.3600	2.87922	-0.033
0.7000	0.78533	-0.3844	3.11228	0.028
0.8500	0.78068	-0.3134	3.26634	0.014
1.0000	0.77412	0.0000	3.42040	0.000

Table 3 : Densities, vicosites, excess molar volumes V^E, and excess viscosites η^E for mixtures of (x) iso-butanol + (1-x) n-butanol at 303.15K.

X	ρ ($g\ cm^{-3}$)	V^E ($cm^3\ mole^{-1}$)	η (cp)	η^E(cp)
0.0000	0.80153	0.0000	2.30000	0.000
0.1447	0.80042	-0.0150	2.28372	-0.0900
0.3004	0.79926	-0.0252	2.34143	-0.1060
0.4518	0.79809	-0.0325	2.42914	-0.0920
0.5503	0.79728	-0.0348	2.49328	-0.0770
0.7006	0.79605	-0.0349	2.58900	-0.0550
0.8503	0.79472	-0.0230	2.68972	-0.0280
1.0000	0.79330	0.0000	2.79143	0.0000

Table 4 : Densities, vicosites, excess molar volume V^E, and excess viscosites η^E for mixtures of (x) sec-butanol + (1-x) n-butanol at 303.15K.

X	ρ ($g\ cm^{-3}$)	V^E ($cm^3\ mole^{-1}$)	η (cp)	η^E(cp)
0.0000	0.80153	0.0000	2.30000	0.000
0.1503	0.80131	-0.0135	2.30154	-0.029
0.3002	0.80106	-0.0230	2.31508	-0.046
0.4501	0.80077	-0.0284	2.334062	-0.051
0.5502	0.80054	-0.0270	2.36498	-0.047
0.7002	0.80016	-0.0225	2.40952	-0.033
0.8502	0.79977	-0.0158	2.45606	-0.017
1.0000	0.79930	0.0000	2.50360	0.0000

The V^E data obtained here for binary mixtures of n-Butanol, isomer of butanol indicate that there is change in "free volume" in the mixture compared with that in the pure components and the interstital accommodation of isomer of butanol in n-butanol structure lead to a more compact structure and to an observed decrease (negative values) in the excess molar volumes.

The negative deviations observed are attributed to the effect of the hydrogen bond structure and this leads to a decrease in the excess molar volume. The more negative values of V^E for the mixture of tret-butanol + n-butanol than mixtures of iso-butanol + n-butanol and sec-butanol + n-butanol as from figure (1) are a consequence of a more interaction due to the both smaller chain length and geometrical shapes of tert-butanol than iso-butanol and sec butanol which increasing the extent of Hydrogen boding in the system. As the chain length of isomer of butanol icreased the values of V^E becomes less negative and the positive

effect becomes more important than the negative. This behavior is similar to that reported for various alkanols and n-alkanes[12]. The interaction sequence in mixtures, tert-butanol + n-butanol > iso-butanol + n-butanol + n-butanol > sec-butanol + n-butanol.

Excess viscosities η^E are shown, figure (2), with negative lobes indicate the increase interaction on mixing the two alcohol's, leading to a decrease in the viscosity of the binary mixture. Figure (2a), with positive lobe at high mole fraction of tret-butanol indicates that no molecular interaction occur or no hydrogen bounding interaction due to high concentration of tert-butanol and high steric effect of the three methyl groups which reduce the extent of the hydrogen bonding. The values of V^E reflect the molecular interaction while the excess viscosities affected by both, the tength and the geometrical shape of the molecvle, that is why the minimum values of V^E and η^E appear at two different mole fractions.

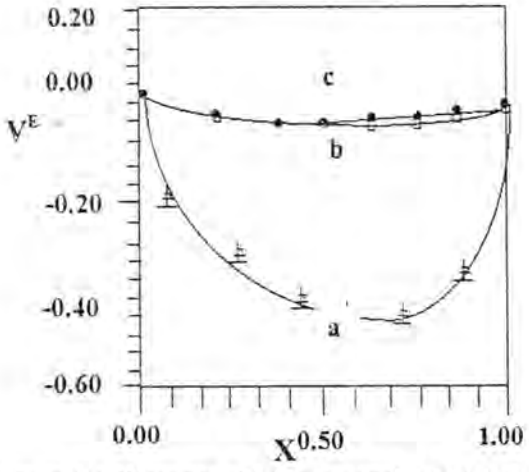

Figure 1 : Mixtures of (a) n-butanol + tert-butanol, (b) n-butanol + iso-butanol and (c) n-butanol + sec-butanol (at 303.15K).

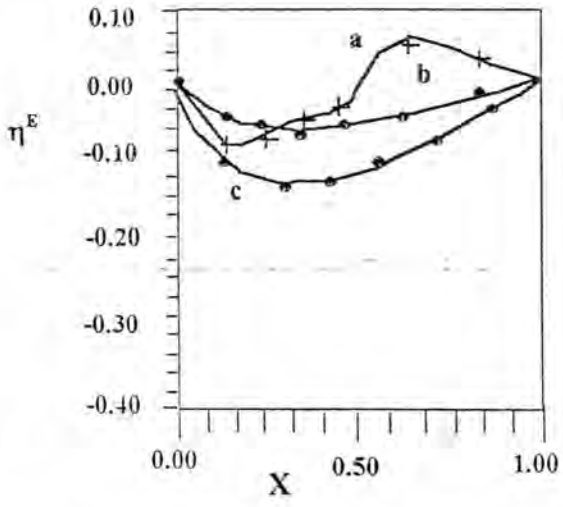

Figure 2 : Mixtures of (a) n-butanol + tert-butanol, (b) n-butanol + iso-butanol and (c) n-butanol + sec-butanol)at 303.15K).

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Reaction of Dimethyl Methoxymethylene Malonate with Some Various Nitrogen Compounds.

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الخلاصة

تضمن البحث دراسة تفاعلات ميثوكسي مثيلين مالونات ثنائي مثيل مع امينو بيردين ، الهيدرازين المائي ، هيدروكسيل امين هيدروكلوريد واورثوفنيلين ثنائي امين . وقد تم تشخيص التراكيب الكيميائية للنواتج على اساس تحليل العناصر الدقيقة لها وخواصها الطيفية .

ABSTRACT

The reaction of dimethylmethoxymethylene malonate with 2-aminopyridine, hydrazine hydrate, hydroxylamine hydrochloride and O-phenylenediamine are reported. Spectral and analytical data were used to confirm the products structure.

INTRODUCTION

Pyrido (1,2-a) pyrimin-4-ones, isooxazolines and pyrazol-5-ones played a vital role in many biological processes[1-5] and their wide applications as plant growth regulators, dyes and corresion inhibitors[6].

Some reactions of dimethyl methoxyomethylene malonate have been reported previously of above anologous heterocyclic ring systems of biological interest[7].

In conjuction with our studies of the reaction of dimethyl methoxymethylene malonate compounds[8], we thought it might be of interest to extend this work to the reactions dimethyl methoxymethylene malonate with some various nitrogen compounds.

RESULTS AND DISCUSSION

2-Aminopyridine condensed easily with ester[1] in the presence of polyphosphoric acid (PPA) giving a quantitative yield of 4H-pyrido (1,2-a) pyrimidin-4-one(II) Scheme1.

Isooxozolone(III), pyrzol-5-one(IV) and dimethyl β-(O-aminoanilino) methylene malonate(V) were obtained respectively by heating the corresponding amino compounds e.g. hydrzime hydrate, hydroxylamine hydrochloride and O-phenylenediamine with alcoholic solution of the ester (1) for 3-5 hours.

The structure of the sythesized compounds (I-V) have been identified by elemental analysis and spectral data (IR, UV[1], HNMR, ^{13}CNMR).

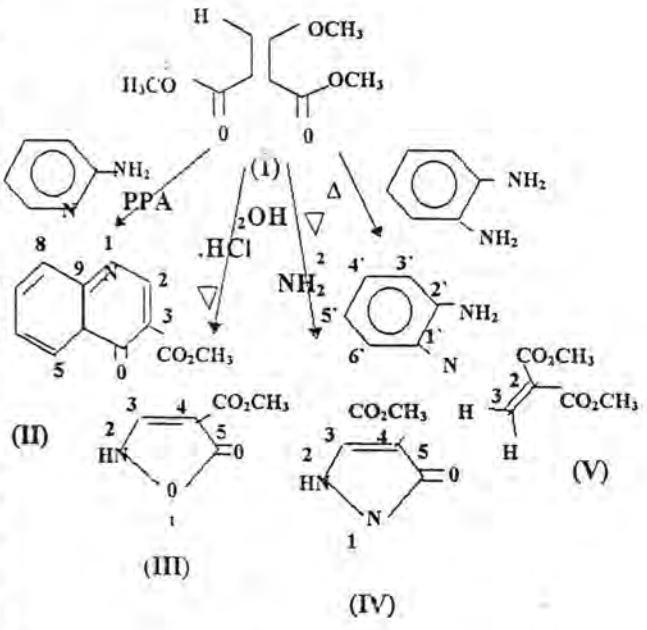
The spectra data provide evidence in support of structure (II-V) for these compounds in which characteristic bands in the regions 1720-1745 cm^{-1} and 1580-1650 cm^{-1} which are attributed to ester

carbonyl group and C=C together with the C=N vibrations respectively, while the C-O stretching vibrations appeared in the region 1640-1680 cm^{-1} as strong bands. The NH bending vibrations for compounds (II-V) appeated in the region 1580-1590 cm^{-1} are attributed to the NH defomation coupled with C=C stretching vibrations. Furthermore, their spectra of the compounds (II-IV) showed two bands in the region 1155-1175 cm^{-1} and 1250-1300 cm^{-1} attributable to C-O-C symmetrical and asymmetriccal stretching vibrations. All the spectra displaced bands in the region 3290-3400 cm^{-1} attributed to NH stretching vibrations.

The ^{1}HNMR data showed a singlet at δ 2.20-4.00 which was integrated for three protons, this was assigned to methyl ester group in sythesised compounds. A singlet at δ 7.30-9.09 expect a doublet in compound (V), integrated for one proton, was assigned to vinylic proton at position 2 or 3 with respect to compounds (I-V). Furthermore, the presence of aromatic and NH or NH_2 protons espacially in compounds (III) and (V) was established by the signals δ 6.75-8.10 and δ 7.50,3.68,10.88 respectively.

The carbon-13 NMR spectra of compounds (II-V) showed the right number of carbon-13 signals. Single frquency off-resonance as well as the known chemical shift rules[9] were used to confirm the assignments of the carbon atoms. FOr the compound (II), the most low field signals at δ159.4 ppm which showed unresolved triplet (long range coupling ^{2}J) ia assigned for C-2. The carbonyl carbon showed signals at δ 165.3 ppm δ 153.6 showed unresolved long range coupling is assigned to C-9 which deshielded by two α-nitrogen atoms(sp^2 and sp^3 nitrogen). The shielded sp^2 carbon at 105.2 ppm was assigned to C-3, this shielding is caused by β-nitrogen effect. Similarly C-8 was assigned for δ128.9 ppm , C-6 was assigned to δ 117.0 ppm. The remaining signals at δ126.9 and δ 139.3 ppm were assigned for C-5 and C-7 respectively.

Similarly, ^{13}C NMR of the remaining compounds (III-V) were investigated and supported the proposed structures (III-V) (see experimental section).



Scheme 1

EXPERIMENTAL

All melting points were determined with a kofler apparatus and uncorrected. IR for Nujol mulls were obtained with Pye Unicam SP-2000 spectrophotometer and the UV spectra were recorded in methanol on Pye Unicam SP 8-20 spectrophotometer. ^{1}H and ^{13}C-NMR spectra were recorded on a Brucker WH 90 DS spectrophotomer equipped with ASPECT 2000,32K computer operated at 22.63 MHZ for ^{13}C-NMR with deterium internal loke. Analyses of the compounds were carried on a CHN analyser, type 1106 (Carbo Erba).

Dimethyl methoxymethylene malonate (1) :

A mixture of dimethyl malonate (153 mmol) and freshly distilled ethyl formate (470 mmol) was added to sodium methoxide (prepared from 160 mmol) of sodium and 150 ml of methanol) and heated at reflux for 4 hours. then evaporated to dryness. The resdue was suspended in dry ether (100 ml) and dimethyl sulphate (155 mmol) was then added slowly and the reaction mixture stirred at room temperture for 48 hours and filtered. The precipitate was washed with ether, filtered and the filtrates evaporated to give brown crystals. Recrystallization from pentane-ether gave compound (1) as white crystalline solids (55%) (m.p. 34-36 °C) V_{max}, 1740 (C=O), 1650 (br, C=O), 1595 (C=C), 1180 cm^{-1} (C-O). ^{1}HNMR ($CDCl_3$), 3.61 (s, 6H, OCH_3), 8.5 (s, 1H, vinyl).

4H-Pyrido (1,2-a) pyrimidin -4- one (!!):

A xiture of 2-aminopridine (25 mmol), ester (1) (25 mmol) and six fold excess (by weight) of polyphosphoric acid was heated at steam bath temperture for 3 hours with mechanical strring. The reaction product was poured into ice-water mixture and the light solid formed was collected, washed with cold and dried at 60 °C. Recrystallization from ethanol gave (92%) yield of the title compound (II) as white crystals m.p. 156-158 °C. (Found, C, 57.9; H, 3.7; N, 13.4 requires C, 58.8; H, 3.9; N, 13.7%); V_{max}, 1720 (C=O), 1640 (C=C), 1580 cm^{-1}(C=C, C=N); λ_{max} (MeOH), 255 and 268 nm; ^{1}HNMR(DMSO-d_6), δ_H, 9.09(s, 1H, 2-H), 9.3 (dd, J=9.0, 2.0, 1H, 5-H), 7.8-8.1 (m, 2H, 6-H and 7-H), 7.5 (dd, J=9.0, 1.2, 1H, 8-H), 4.0 (s, 3H, OCH_3); δ_C, 159.4 (C-2), 105.2 (C-3), 15.47 (C-4), 126.9 (C-5), 117 (C-6), 139.3 (C-7), 128.9 (C-8), 153.6 (C=N), 165.3 ($COOCH_3$(, 52.1 ($COOCH_3$).

4-Methoxycarbonyl isoxazolin-5-one (III) :

A solution of NH_2OH. HCl(0.15 mol) in water (15 ml) wasadded to the solution of the ecter (1) (0.1 mol) in mathanol (100 ml) and the mixture was heated to reflux for 1.5 hr. The residue from the solvent evaporation was taken up in water (80 ml) and after cooling in ice, the isoxazolone (III0 was filtered off and recrystallized from ethanol 76%.m.p. 186-188 °C (found, C, 41, H, 3.62; N, 9.44 requires C, 41.96; H, 3.50; N, 9.79%); V_{max}, 1745, 1728 (C=O), 1650 (C=C), 1580, 1550 (δNH) 3300-3250 cm^{-1} (NH); λmax(MeOH), 283, and 268 nm; ^{1}HNMR (DMSO-d_6); δ_H, 5.3 (s, 1H, 3-H), 2.2 (s, 3H, OCH_3), 7.5 (s, 1H, NH), δ_C 151.9 (C-4), 85.7 (C-3), 170.5 (C-5), 162.3 ($COOCH_3$), 50.8 ($COOCH_3$).

4-Methoxycarbonyl pyrazol -5- one (IV) :

A mixture of ester (1) (25 mmol) and hydrazine hydrate 99-100% (30 mmol) in 20 ml ofabsolute ethanol was refluxed for 3hrs. Removal of the solvent and excess of reagent under reduced pressure gave yellow-orange residue. Recystaliza -tion from petroleum ether gave 72& of compound (IV) .m.p. 244-246 °C (found, C, 41.96; H, 4.45; N, 19.84 requies C, 42.25; H, 4.23; N, 19.72%); V_{max}, 3380-3400 (NH), 1730, 1680 (C=O), 1550 (δ N-H), 1365 (C-N), 1600, 1590 cm^{-1} (C=C), λmax(MeOH), 256 and 285 nm; ^{1}HNMR(DMSO-d_6); δH 7.3(s, 1H, 3-H), 3.65 (s, 3H, OCH_3); δC 91.1 (C-3), 139.5 (C-4); 165.0 (C-5), 164.7 ($COOCh_3$), 49.3 ($COOCH_3$).

Dimethyl β-(o-aminoanilino) methy -lene malonate (V) :

A mixture of ester (1) (25 mmol) and o-phenylene diamine (25 mmol) in 20 ml of absolute ethanol was refluxed for 5hrs. Evaporation of solvent gave pale yellow residue. recrystallization from ethanol (Charcoal) gave 86% of compound (V) as light yellow crystals m.p.

222-224 °C (Found, C, 56.86, H, 5.12; N, 11.64 requires C, 57.60; H, 5.69; N, 11.20%), V_{max}, 1720, 1680 (C=O), 1645, 1600 (C=C), 1560 (δ NH), 1500 (C=N), 3290 cm^{-1}; λmax(MeOH), 295 and 338 nm; ^{1}HNMR ($CDCl_3$); δ H, 3.68(s, 2H, NH_2), 3.75 and 3.85(s, 6H, $2OCH_3$), 6.75 (m, 4H, aromatic), 8.45 (dd, 1H, vinylic), 10.88(dd, 1H, NH), δC 51.3 and 51.5 ($2COOCH_3$), 92.9(vinylic =Ch-), 154.6 (HN-CH=), 127.8 (C-1`), 126.7 (C-2`) 177.8(C-3`), 120.1 (C-4`), 138.2 (C-5`), 119.0 (C-6`).

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Geometric Aspects of Four Minor Forms of The Desargues Proposition

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الخلاصة

يتضمن هذا البحث دراسة للعلاقات بين قظايا صفريات ديسارك وصفريات ديسارك الثنوية التشكلية وامكن اثبات ان قضايا صفريات ديسارك تكافئ قضايا صفريات ديسارك الثنوية اذا وفقط اذا كانت p′ و L′ تمتلكان نفس العدد من العناصر حيث ان P′ تمثل المجموعة التي كل عنصر من عناصرها هي النقطة الرابعة على ذلك المستقيم و L′ هي المجموعة التي كل عنصر من عناصرها يمثل المستقيم الذي تقع عليه اربعة نقاط . كذلك امكن التوصل الى ان L′ تمتلك على الاكثر اربعة عناصر وان أي ثلاثة عناصر من L′ لاتلتقي في نقطة واحدة ، وبذلك تمكنا من تحديد عدد صفريات ديسارك الثنوية .

ABSTRACT

The purpose of this paper is to study the Geometrical properties of four minor forms of the Desargues proposition. It is found that all minor Desargues and their duals are equivalent if and only if, P′ and L′ has the same number of element.

INTRODUCTION

Let (1,2,3)and (1′,2′,3′) be any two triangles in a projective plane π. If [11′], [22′] and [33′] are concurrent, then the two triangles are said to be perspective from a point. In symbols,
O=[11′]∩[22′]∩[33′].
If [12]∩[1′2′]=3″,[13]∩[1′3′]=2″
and [23]∩[2′3′]=1″
form a collinear triple, denoted by (1″,2″,3″) the two triangles are said to be perspective from the line [1″2″]. Hence the proposition of Desargues, denoted by D, may be stated as follows :

If two triangles are perspective from a point, they are perspective from a line, and their dual: If two triangles are perspective from a line, they are perspective from a point.

MINOR FORMS OF THE DESARGUES PROPOSITION.

Let P={1,2,3,1′,2′,3′,1″,2″,3″,0} be the set of points and
L={[1″],[2″],[3″],[0],[1],[2],[3],[1′],
[2′],[3′]}
be the set of line then :
Simple minor Desargues
D.⇔(∃!i)(i∈P)({I[i])([i]∈L)}).
other minor forms of Desargues, may be formulated as follows:
minor Desargues $D_{P'}$⇔∃P′⊆P such that iI[i} for each i∈P′ and [i]∈L.

If P′ has two elements them we called : double simple Desargues.
$D_{[i,j]}$ where i≠j and j[i]. (P′ represented {i,j})
If P′ has three elements then we called : triple simple Desargues .
$D_{[i,j,k]}$ where i≠j≠k≠i and iI[i],kI[i]. (P′ represented {i,j,k})
If p′ has four elements then we called :quadruple simple Desargues.
$D_{[i,j,k,h]}$ where i,j,k,h are different and iI[i], kI[i], hi[i]. (P′ represented {i,j,k,h})(see Table 1).

Table 1

O	2′	3	2	3′	1	2″	3″	1′	1″
1	O	3′	1″	2′	3	1′	2	3″	2″
1′	2	O	3	1″	2″	3′	1	2′	3″
[1″]	[2″]	[3″]	[1′]	[1]	[2′]	[2]	[3′]	[3]	[O]
1″	2″	3″	1′	1	2′	2	3′	3	O

DUAL DESARGUES;

If two triangles are perspective from a line, they are perspective from a point.
Simple minor dual Desargues
$\tilde{D}_{[i]} \Leftrightarrow (\exists!i)(i\in P)(iI[i])([i]\in L)$.

Other minor forms of dual Desargues, may be formulated as follows:
Minor dual Desargues
$D_{L'} \Leftrightarrow \exists L' \subseteq L$ such that iI[i] for each $[i]\in L'$ and $i\in P$. (see Table 2).

THEOREM 1

In minor dual Desargues $\tilde{D}_{L'}$, L′ has at most four elements such that no three of which are concurrent.

PROOF

If L′ is an empty set then we get dual Desargues proposition. Another cases L′ has at most four elements since, if L′ has more than four elements we get at least two distinct lines that are incident with two points. Which is a contradiction by axiom (i) and (ii) of projective plane.
Illustration, if L′={([1],[1′],[2″],[3″], [O]} then we get the line
[O]∩[2″]={O,2″}also[O]∩[3″]={O,3″}

Now if L′ has four elements no three of which are concurrent also, if L′ has three diagonal lines of a complete quadrilateral are not concurrent. We illustrate by the following example;
If L′={[1],[2″],[3″]} we remark that the elements [1],[2″],[3″] represented the diagonal line of a complete quadrilateral [2′] [O][3′][1′].

COROLLARY 1

There exist only 65 minor dual Desargues.

PROOF

Type I: Simple dual Desargues. There exist only ten simple dual Desargues is as follows:
$\tilde{D}_{[i]} \Leftrightarrow (\exists!i)\ (i\in P)\ (iI[i])\ ([i]\in L)$,
Also, we have the following type obtained from definition of minor $\tilde{D}$, theorem 1 and axiom (i), (ii) of projective plane.

Type II : Double simple dual Desargues.
$\tilde{D}_{[[i],[j]]}$ where i≠j [i], iI[i], jI[j], [i]≠[j] with i,j∈P and [i],[j]∈L′(L′ represented {[i],[j]}).

Type III: Triple simple dual Desargues:
$\tilde{D}_{[[i],[j],[k]]}$ where i≠j≠k≠i, [i]≠[j]≠[k]≠[i] and j[i], k[i], iI[i], jI[j], kI[k] (L′ represented {[i],[j],[k]}).

Type IV: quadruple simple dual Desargues :

$\widetilde{D}_{\{[i],[j],[k],[h]\}}$ where i,j,k,h are different and [i],[j],[k],[h] are different and iI[i],jI[j],kI[k],hI[h],j[i],k[i],h[i].
(L′ represented {[i],[j],[k],[h]}). (see Table 2).

Table 2

[O]	[2′]	[3]	[2]	[3′]	[1]	[2″]	[3″]	[1′]	[1″]
[1]	[O]	[3′]	[1″]	[2′]	[3]	[1′]	[2]	[3″]	[2″]
[1′]	[2]	[O]	[3]	[1″]	[2″]	[3′]	[1]	[2′]	[3″]
1″	2″	3″	1′	1	2′	2	3′	3	O
[1″]	[2″]	[3″]	[1′]	[1]	[2′]	[2]	[3′]	[3]	[O]

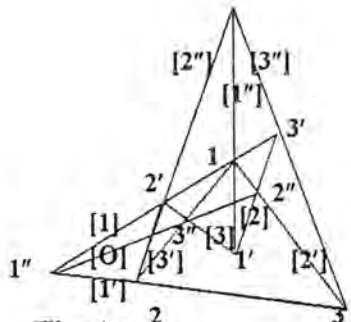

Fig. 1.

Now, we relate the Desargues proposition to their duels.

THEOREM 2.

D_1 is equivalent to its dual

PROOF

We have only to show that D_1 implies its dual $\widetilde{D}_{[i]}$.

This states that for 7 lines [O],[1],[2],[3],[1′],[2′],[3′], with [1]∩[1′],[2]∩[2′],[3]∩[3′]I[O], [3′]∩[2′] I[1] that the lines
[1″]=[([3′]∩[2′])([3]∩[2])],
[2″]=[([3]∩[1])([3′]∩[1′])],
[3″]=[([1]∩[2])([1′]∩[2∩])]
are concurrent.
Defining the points 1,2,3,1′,2′,3′,1″, 2″,3″ as in Fig 1, we will transform $\widetilde{D}_{[i]}$ in to the converse of D_1.
If 1″,2″,3″ are collinear them [11′], [22′], [33′], are concurrent. Now to derive this from D_1, we define
O=[33′]∩[22′], i‴=[O1]∩[2′3″]
and get, upon applying D_1 to the two triangles (1,2,3) and (1‴,2′,3′),
2″=([O]∩{13]) I[1‴3′], 1‴I[2″3′]
=[1′3′] and further
1‴=[1′3′]∩[2′3″]=1′ which proves, the desires conclusion, Oi[11′]

THEOREM 3.

All simple minor dual Desargues are equivalent. proof

We prove this theorem by changing [i] of $\widetilde{D}_{[i]}$ with respect to $\widetilde{D}_{[i]} \Leftrightarrow$ (∃! [i]) ([i] εL) (i][i]) iεP).

Now, a consequence of Theorem 2, 3 is the following theorem.

THEOREM 4.

All simple minor Desargues and their duels are equivalent.

THEOREM 5.

$D_{[1,1']}$ is equivalent to its dial.

PROOF

We have only to show that $D_{[1,1']}$ implies its dual $\widetilde{D}_{\{[1],[1']\}}$.This states that for 7 lines [O],[1],[2],[3], [1′],[2′],[3′] with [1]∩[1′],[2]∩[2′], [3]∩[3′] I[O],[2]∩[3]I[1′] and [2′]∩[3′]I[1] that the lines
[1″]=[([2′]∩[3′])([2]∩[3])],
[2″]=[(3′]∩[1′])([3]∩[1])] and
[3″]=[([1′]∩[2′])([1]∩[2])] are concurrent.
Defining the points 1,2,3, 1′,2′,3′,1″.2″,3″ as in Fig 2, we will transform $\widetilde{D}_{\{[1],[1']\}}$ in to the converse of $D_{\{[1,1']\}}$. If 1″,2″,3″ are collinear then [11′],[22′],[33′] are concurrent. Now, to derive this from $D_{[1,1']}$, we define
O=[33′]∩[22′],

$1'''=[O1]\cap[2'3'']$ and get, upon applying $D_{\{1,1'\}}$ to the two triangles (1,2,3) and $(1''',2',3')$, $2''=([O]\cap[13])I[1'''3']$, $1'''I[2''3']==[1'3']$ and further $1'''=[1'2']\cap[2'3'']=1'$ which proves the desired conclusion, $Oi[11']$.

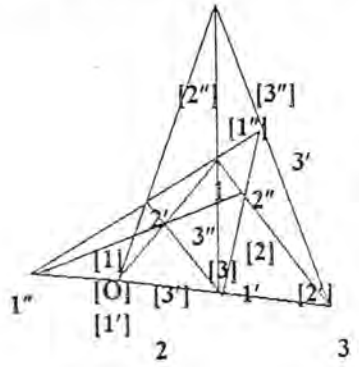

Fig. 2

THEOREM 6

All double simple of minor dual disargues are pair-wise equivalent.

PROOF

We prove this theorem by changing [i] or [j] or both of them of $\widetilde{D}_{\{[i],[j]\}}$ with respect to element of L respectively such that:

$\widetilde{D}_{\{[i],[j]\}}$ where $[i]\neq[j]$ and jI[j], iI[i], i[j] $(i,j\in P$ and $[i],[j]\in L)$. Now, a consequence of Theorem 5,6 is the following theorem.

THEOREM 7

All double simple of minor Desargues and their duels are equivalent.

THEOREM 8

$D_{\{1,2,3\}}$ is equivalent to its dual.

PROOF

We have only to show that $D_{\{1,2,3\}}$ implies its dual $\widetilde{D}_{\{[1],[2],[3]\}}$. This states that for 7 lines [O],[1],[2],[3],[1′],[2′],[3′] with $[1]\cap[1\cap],[2]\cap[2'],[3]\cap[3'][O]$, $[1']\cap[3']I[2],[1']\cap[2\cap]I[3]$ and $[2']\cap[3']I[1]$ that the lines $[1'']=[([3']\cap[2'])([3]\cap[2])]$, $[2'']=[([1]\cap[3])([1']\cap[3'])]$ and $[3'']=[([1]\cap[2])([1']\cap[2'])]$ are concurrent. Defining the points 1,2,3,1′,2′,3′,1″,2″,3″ as in Fig 3, we will transform $\widetilde{D}_{\{[1],[2],[3]\}}$ in to the converse of $D_{\{1,2,3\}}$.

If 1″,2″,3″ are collinear then [11′],[22′],[33′] are concurrent. Now, to derive this from $D_{\{1,2,3\}}$, we define $O=[33']\cap[22']$, $1'''=[O1]\cap[2'3'']$ and get, upon applying $D_{\{1,2,3\}}$ to the two triangles (1,2,3) and $(1''',2',3')$, $2''=([O]\cap[13])I[1'''3']=[1'3']$ and $1'''=[1'3']\cap[2'3'']=1'$ which proves, the desired conclusion, $OI[11']$

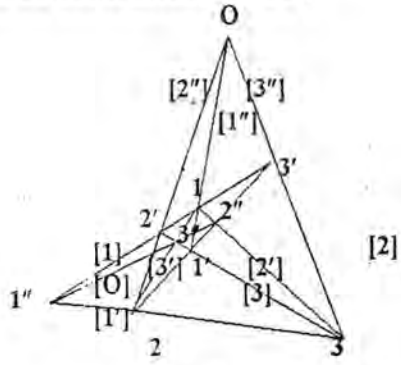

Fig. 3

THEOREM 9

All triple simple of minor dual Desargues are pair-wise equivalent.

PROOF

We prove this theorem by changing [i] or [j] or [k] or any two of them or [i] and [j] and [k] of $\widetilde{D}_{\{[i],[j],[k]\}}$ with respect to element of L respectively such that: $\widetilde{D}_{\{[i],[j],[k]\}}$. where $[i]\neq[j]\neq[k]\neq[i]$ and j[i], with iI[i], jI[i] and Ki[k] $(i,j,k\in P$ and $[i],[j],[k]\in L)$. Now, a consequence of Theorem 8,9 is the following theorem.

THEOREM 10

All triple simple of minor Desargues and their duels are equivalent.

THEOREM 11

$D_{\{1,2,3,O\}}$ is equivalent to its dual.

PROOF

We have only to show that $D_{\{1,2,3,O\}}$ implies its dual $\tilde{D}_{\{[1],[2],[3],[O]\}}$. This states that for 7 lines [O],[1],[2],[3],[1′],[2′],[3′] with
[1]∩[1′],[2]∩[2′],[3]∩[3′]I[O],
[1′]∩[3′]I[2],[1′]∩[2′]I[3],
[2′]∩[3′]I[1] and [2″]∩[1″]∩[3″]I[O] that the lines
[1″]=[([2′]∩[3′])([2]∩[3])],
[2″]=[([3′]∩[1′])([3]∩[1])] and
[3″]=[([1′]∩[2′])([1]∩[2])] are concurrent. Defining the points 1,2,3,1′,2′,3′,
1″,2″,3″ as in Fig 4, we will transform $\tilde{D}_{\{[1],[2],[3],[O]\}}$ in to the converse of $D_{\{1,2,3,O\}}$. If 1″,2″,3″ are collinear then[11′],[22′],[33′] are concurrent. Now, to derive this from $D_{\{1,2,3,O\}}$, we define O=[33′]∩[22′],
1‴=[O1]∩[2′3″] and get, upon applying $D_{\{1,2,3,O\}}$ to the two triangles (1,2,3) and (1‴,2′,3′),
2″=([O]∩[13])I[1‴3″], 1‴I[2″3′=[1′3′] and further
1‴=[1′3′]∩[2′3″]=1′ which proves the desired conclusion OI[11′].

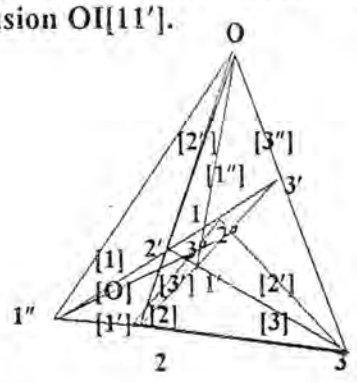

Fig. 4

THEOREM 12

All quadruple simple of minor dual Desargues are pair-wise equivalent.

PROOF

We prove this theorem by changing three elements from {[i],[j],[k],[h]} or all these elements of $\tilde{D}_{\{[i],[j],[k],[h]\}}$ with respect to element of L respectively such that:
$\tilde{D}_{\{[i],[j],[k],[h]\}}$ where [i],[j],[k],[h] are different and i,k,h[i] with iI[i],jI[j], kI[k] and hI[h] 9i,j,k,h∈P and [i],[j], [k],[h]∈L). Now, a consequence of theorem 11,12 is the following theorem.

THEOREM 13

All quadruple simple of minor Desargues and their duels are equivalent .

Now, a consequence of theorems 4,7,10,13 is the following theorem.

THEOREM 14

All minor Desargues and their duels are equivalent if P′ and L′ has the same number of element.

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Infinite General Prolate Spheroidal Transform of Several Variables

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الخلاصة

هدف هذا البحث هو ايجاد تعميم غير منتهي لتحويل كروي متطاول يتضمن دوال لعدة متغيرات . كذلك ايجاد الصيغة العكسية لهذا التحويل ، وهذه الصيغة طبقت بمثال .

ABSTRACT

The aim of this paper is to investigate an infiinite generalized prolate spheroidal transform involving functions of several variables. An invertion formula for this transform has been established and this formula has been varified by an example.

INTRODUCTION

R.K. Gopta (1977) defined generalized prolate spheroidal wave functions by[1]

$$\psi_{Nn}(ys)=(1/V_{Nn})\sum_{j=0}^{\infty} d_j^{(Nn)} \frac{J_{N+2j+1}(ys)}{\left(\frac{N+j}{j}\right)\sqrt{ys}} \quad ...(1)$$

where $J_{N+2\,j+1}(ys)$ is the Bessel function of first kind .

The Hankel transform of two variables has been defined by Chauhan (1985) as [2]

$$f(s_1,s_2)=\int_0^{\infty}\int_0^{\infty} x_1x_2 f(x_1,x_2)J_{n1}(x_1s_1)J_{n2}(x_2s_2)dx_1dx_2 \quad ...(2$$

In this paper, our aim is to generalize this transform by investigationg infinite generalized prolate spheroidal transform involving functions of several variables.

DEFINITION

The infinite generalized prolate spheroidal transform of several variables is defined by the integral equation

$$\phi(s_1,s_2,...,s_n)=\prod_{k=1}^{n}\eta(s_k)\int_0^{\infty}\int_0^{\infty}...\int_0^{\infty} f(y_1,y_2,...,y_n)\prod_{k=1}^{n}\eta\psi_{N_k,n_k}(y_ks_k)dy_1dy_2...dy_n...(3$$

where $\prod_{k=1}^{n}\eta\,\psi_{N_k,n_k}(y_ks_k)$, k=1,2,...,n are the generalized prolate spheroidal wave functions defined by (1) and the function $f(y_1, y_2, ...,y_n)$ and the parameters are so restricted that the multiple integral[3] is absolutely convergevt.

INVERSION THEORY

If the infinite generalized prolate spheroidal transform of several variables is defines by (3) then

$$(1/2)[f((y_1, y_2, ..., y_n)+ O) + f((y_1, y_2, ...,y_n) - O)] =$$

$$\frac{1}{(2\pi i)^n}\int_{c-i_\infty}^{c+i_\infty}\int_{c-i_\infty}^{c+i_\infty}...\int_{c-i_\infty}^{c+i_\infty}\left[\frac{2^{\sum_{k=1}^{n}p_k+1}\prod_{k=1}^{n}\eta(V_{N_k,n_k}y_k^{-p_k}}{N_k,n_k}\right]$$

$$\sum_{j_1=0}^{\infty}\sum_{j_2=0}^{\infty}.....\sum_{j_n=0}^{\infty}\prod_{k=1}^{n}\left\{\frac{d_{j_k}\Gamma(3/4-p_k/2+N_k/2+j_k)}{\left(\frac{N_k+j_k}{j_k}\right)\Gamma(5/4+N_k/2+j_k+p_k/2)}\right\}$$

$$dp_1dp_2...dp_n,....(4$$

PROOF

Substituting the values of $\psi_{N_k,n_k}(y_k s_k)$, k=1,2,...,n from (1) in the intergal equation (3) we get

$$\phi(s_1,s_2,...,s_n)=\prod_{k=1}^{n}\eta(s_k)\int_0^\infty\int_0^\infty\cdots\int_0^\infty\frac{f(y_1,y_2,...,y_n)}{\prod_{k=1}^{n}V_{N_k,n_k}}\sum_{j_1=0}^{\infty}\sum_{j_2=0}^{\infty}\cdots\sum_{j_n=0}^{\infty}$$

$$\left[\prod_{k=1}^{n}d_{j_k}^{(N_k,n_k)}\frac{J_{N_k+2j_k+1}(y_k s_k)}{\binom{N_k+j_k}{j_k}\sqrt{y_k s_k}}\right]dy_1dy_2\cdots dy_n,\quad\ldots\ldots\ldots(5)$$

where the multiple integrals (5) is absolutly convergent.

Now we evaluate

$$I=\int_0^\infty\int_0^\infty\cdots\int_0^\infty\prod_{k=1}^{n}(s_k^{-p_k-1})\phi(s_1,s_2,...,s_n)ds_1ds_2\cdots ds_n$$

$$=\int_0^\infty\int_0^\infty\cdots\int_0^\infty\prod_{k=1}^{n}(s_k^{-p_k-1})\left(\prod_{k=1}^{n}(s_{k=1})\right)\int_0^\infty\int_0^\infty\cdots\int_0^\infty\frac{f(y_1,y_2,..y_n)}{\prod_{k=1}^{n}V_{N_k,n_k}}$$

$$\sum_{j_1=0}^{\infty}\sum_{j_2=0}^{\infty}\cdots\sum_{j_n=0}^{\infty}\prod_{k=1}^{n}\left(d_{j_k}^{(N_k,n_k)}\frac{J_{N_k+2j_k+1}(y_k s_k)}{\binom{N_k+j_k}{j_k}\sqrt{y_k s_k}}\right)(dy_1dy_2..dy_n)$$

$$ds_1ds_2\ldots ds_n,\quad\ldots\ldots\ldots(6)$$

substituting $y_k s_k = z_k$ (k=1,2,...,n), we have

$$I=\int_0^\infty\int_0^\infty\cdots\int_0^\infty\prod_{k=1}^{n}(s_k^{-p_k-1})f(y_1,y_2,...,y_n)\int_0^\infty\int_0^\infty\cdots\int_0^\infty\frac{\prod_{k=1}^{n}(z_k^{-p_k})}{\prod_{k=1}^{n}(V_{N_k,n_k})}$$

$$\sum_{j_1=0}^{\infty}\sum_{j_2=0}^{\infty}\cdots\sum_{j_1=0}^{\infty}\prod_{k=1}^{n}\left(d_{j_k}^{(N_k,n_k)}\frac{J_{N_k+2j_k+1}(z_k)}{\binom{N_k+j_k}{j_k}\sqrt{z_k}}\right)dz_1dz_2\cdots dz_n)dy_1dy_2..dy_n$$

$$=\left(\prod_{k=1}^{n}V_{N_k,n_k}\right)^{-1}\sum_{j_1=0}^{\infty}\sum_{j_2=0}^{\infty}\cdots\sum_{j_1=0}^{\infty}\prod_{k=1}^{n}\left[\frac{d_{j_k}^{(N_k,n_k)}}{\binom{N_k+j_k}{j_k}}\right]\int_0^\infty\int_0^\infty\cdots\int_0^\infty y_k^{p_k-1}f(y_1,y_2,...,y_n)$$

$$dy_1,dy_2,\ldots,dy_n\int_0^\infty\int_0^\infty\cdots\int_0^\infty\prod_{k=1}^{n}|z_k^{-p-1/2}J_{N_k+2j_k+1}(z_k)|dz_1dz_2\ldots dz_n\ldots..(7)$$

Term by term integration is justified due to uniform cinvergence of the generalized prolate spheroidal wave functions. The integral

$$\int_0^\infty\int_0^\infty\cdots\int_0^\infty\prod_{k=1}^{n}[z_k^{-p-1/2}J_{N_k+2j_k+1}(z_k)]dz_1dz_2\ldots dz_n$$

is convergent now we let

$$1,p_2,\ldots,p_n)=$$

$$\int_0^\infty\int_0^\infty\cdots\int_0^\infty\prod_{k=1}^{n}(s_k)^{-p_k-1}\phi(s_1,s_2,\ldots,s_n)ds_1ds_2\ldots ds_n$$

$$=\left(\prod_{k=1}^{n}V_{N_k,n_k}\right)^{-1}\sum_{j_1=0}^{\infty}\sum_{j_2=0}^{\infty}\cdots\sum_{j_n=0}^{\infty}\prod_{k=1}^{n}\left(d_{j_k}^{N_k,n_k}/\binom{N_k+n_k}{j_k}\right)\int_0^\infty\int_0^\infty\cdots\int_0^\infty\prod_{k=1}^{n}$$

$$(Z^{-p_k-1/2}J_{N_k+2j_k+1}(z_k))dz_1dz_2\ldots dz_n\int_0^\infty\int_0^\infty\cdots\int_0^\infty\prod_{k=1}^{n}(y_k^{p_k-1})$$

$$f(y_1,y_2,\ldots,y_n)dy_1dy_2\ldots dy_n$$

$$=\left(\prod_{k=1}^{n}V_{N_k,n_k}\right)^{-1}2^{-\sum_{k=1}^{n}p_k-1}\sum_{j_1=0}^{\infty}\sum_{j_2=0}^{\infty}\cdots\sum_{j_n=0}^{\infty}\prod_{k=1}^{n}\left(\frac{d_{j_k}^{N_k,n_k}}{\binom{N_k+n_k}{j_k}}\right)\tilde{f}(y_1,y_2,\ldots,y_n)$$

$$\frac{\prod_{k=1}^{n}\Gamma(3/4-p_k/2+N_k/2+j_k)}{\prod_{k=1}^{n}\Gamma(5/4+N_k/2+j_k+p_k)},[-1<p_k<\min(N_{k+2j_k+O/2})k=1,2,..,n]$$

then

$$\tilde{f}(y_1,y_2,..,y_n)=2^{\sum_{k=1}^{n}p_k+1}\prod_{k=1}^{n}(V_{N_k,n_k})F(p_1,p_2,\ldots,p_n)$$

$$\left[\sum_{j_1=0}^{\infty}\sum_{j_2=0}^{\infty}\cdots\sum_{j_n=0}^{\infty}\prod_{k=1}^{n}\left(\frac{d_{j_k}^{N_k,n_k}\Gamma(3/4-p_k/2+N_k/2+j_k)}{\binom{N_k+j_k}{j_k}\Gamma(5/4+N_k/2+j_k+p_k)}\right)\right]^{-1}..(8)$$

By mellion inversion furmula for n-variables [3] the theorem is completely established.

SPECIAL CASES

(I) When N_k=+1/2 (k=1,2,..,n) in the definition (3) and using therelation (1), we get

$$\phi(s_1,s_2,\ldots,s_n)=\prod_{k=1}^{n}(s)_k\int_0^\infty\int_0^\infty\cdots\int_0^\infty\frac{f(y_1,y_2,\ldots y_n)}{(\prod_{k=1}^{n}V_{1/2,n_k})}$$

$$\sum_{j_1=0}^{\infty}\sum_{j_2=0}^{\infty}\cdots\sum_{j_n=0}^{\infty}\prod_{k=1}^{n}[d_{j_k}^{1/2,n_k}\frac{J_{2j+O/2}(y_k s_k)}{\binom{1/2+j_k}{1/2}\sqrt{y_k s_k}}]$$

$$dy_1dy_2\ldots dy_n,\quad\ldots\ldots(9)$$

if we hve replace $f(y_1,y_2,...,y_n)$ by

$$\prod_{k=1}^{n}[\sqrt{(y_k/s_k)^J}.(\frac{V_{1/2,n_k}}{\sum_{k=1}^{n}d_{j_k}^{1/2,n_k}})\binom{1/2+j_k}{1/2}]\,a$$

$f(y_1,y_2,\ldots,y_n)$

nd $\phi(s_1,s_2,...,s_n)$ by $f(s_1,s_2,...,s_n)$ then equation (9) becomes

$$\tilde{f}(s_1,s_2,\ldots,s_n)=\int_0^\infty\int_0^\infty\cdots\int_0^\infty\prod_{k=1}^{n}[y_kJ_{2j_k+O/2}(y_k s_k)]$$

$$f(y_1,y_2,\ldots,y_n).dydy\ldots dy,\ldots(10)$$

which is also a new result called Hankel transform of n-variables. If we take n=2 in (9) we get the result(2).

(II)When $N_k=\pm 1/2$ **and** $\dfrac{d_{j_k}^{N_k,n_k}}{\binom{N_k+j_k}{j_k}} \longrightarrow 0$,

we have

$$=1/2[\int[(y_1,y_2,\ldots,y_n)+O]+\int[(y_1,y_2,\ldots,y_n)-O)]$$

and

$$=1/(2\Pi i)^n \int_{c-i\infty}^{c+i\infty}\int_{c-i\infty}^{c+i\infty}\ldots\int_{c-i\infty}^{c+i\infty} F(p_1,p_2,\ldots,p_n)2^{-\sum_{i=1}^{n}p_k+1}\Pi_{k=1}^{n}$$

$$[y_k^{-p_k}\frac{\Gamma(1-p_k+j_k)}{\Gamma(3/2+p_k+j_k)}]dp_1dp_2\ldots dp_n,\ldots.(11)$$

$$\phi(s_1,s_2,\ldots,s_n)=$$

$$\Pi_{k=1}^{n}(s_k)\int_0^\infty\int_0^\infty\ldots\int_0^\infty\int(y_1,y_2,\ldots,y_n)\Pi_{k=1}^{n}$$

$$[\frac{J_{2j_k+O/2}(y_k s_k)}{\sqrt{y_k s_k}}]dy_1dy_2\ldots dy_n,\ldots.(12)$$

This gives the inversion formula for Hankel transform of n-variables.

EXAMPLE

Let $\int(y_1,y_2,\ldots y_n)=\Pi_{k=1}^{n}(y_k^{-1/2})$

$$\exp\left[-\sum_{k=1}^{n}(a_k/y_k)\right],(a_k>0,k=1,2,\ldots,n)$$

Using equation (9) and mellion's inversion formula of n-variables $,\phi(s_1,s_2,\ldots,s_n)$ **can be easily evaluated.**

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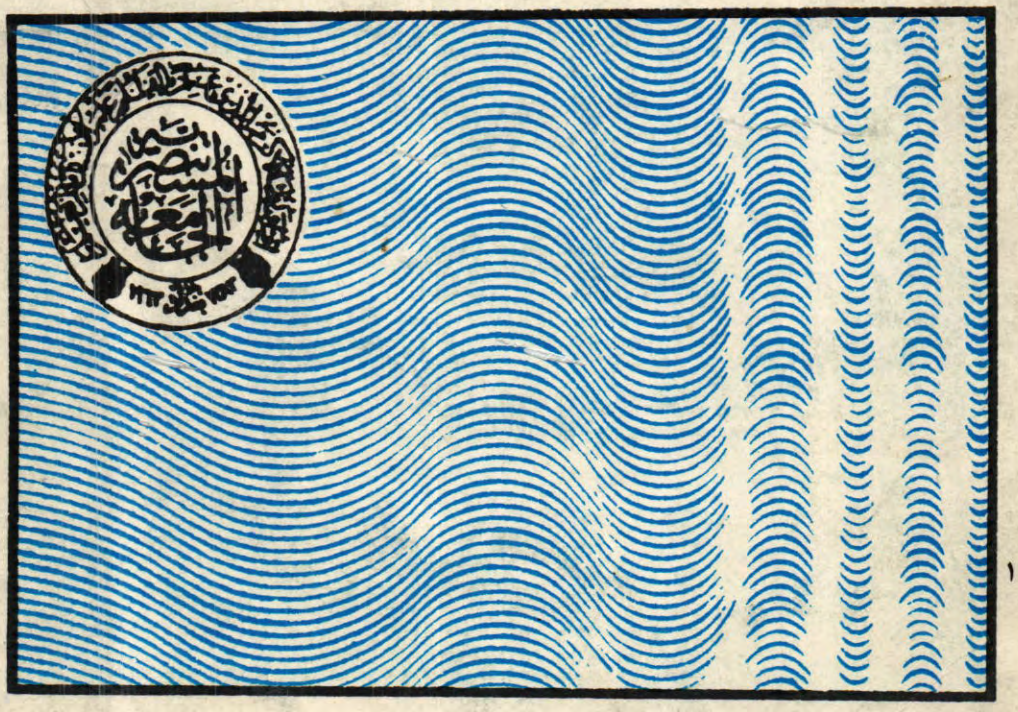

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مجلة علوم المستنصرية

تعليمات النشر

١ـ تقوم المجلة بنشر البحوث الرصينة التي لم يسبق نشرها في مكان اخر بعد اخضاعها للتقويم العلمي من قبل مختصين وباي من اللغتين العربية او الانجليزية .

٢ـ يقدم الباحث او الباحثون طلبا تحريريا لنشر البحث في المجلة على ان يكون مرفقا بثلاث نسخ من البحث مطبوعة على الالة الكاتبة بترك فراغين (double space) بين سطر واخر على ورق ابيض قياس (A4) من النوع الجيد وتترك مسافة (٥ر٢) سم على جانبي كل صفحة .

٣ـ يطبع عنوان البحث واسماء الباحثين (كاملة) وعناوينهم باللغتين العربية والانجليزية على ورقة منفصلة شرط ان لاتكتب اسماء الباحثين وعناوينهم في اي مكان اخر من البحث وتعاد كتابة البحث فقط على الصفحة الاولى من البحث .

٤ـ تكتب اسماء الباحثين كاملة بحروف كبيرة (capital) في حالة استخدام اللغة الانجليزية وكذلك الحروف الاولى فقط من الكلمات (عدا حروف الجر والاضافة) المكونة لعنوان البحث وتكتب عناوين الباحثين بحروف اعتيادية صغيرة (small letters)

٥ـ تقدم خلاصتان وافيتان لكل بحث احداهما بالعربية والاخرى بالانجليزية وتطبع على ورقتين منفصلتين بما لا يزيد على (٢٥٠) كلمة لكل خلاصة .

٦ـ تقدم الرسوم التوضيحية منفصلة عن مسودة البحث وترسم على ورق شفاف (tracing paper) بالحبر الصيني الاسود وترفق ثلاث صور لكل رسم وتكتب المعلومات عنها على ورقة منفصلة ولا يجوز تكرار المعلومات ذاتها في الرسوم والجداول في وقت واحد الا اذا اقتضت ضرورة المناقشة ذلك .

٧ـ يشار الى المصدر برقم يوضع بين قوسين بمستوى السطر نفسه بعد الجملة مباشرة وتطبع المصادر على ورقة منفصلة ويستخدم الاسلوب الدولي المتعارف عليه عند ذكر مختصرات اسماء المجلات.

٨ـ يفضل قدر الامكان تسلسل البحث ليتضمن العناوين الرئيسة الاتية : المقدمة طرائق العمل النتائج والمناقشة الاستنتاجات . المصادر. وتوضع هذه العناوين دون ترقيم في وسط الصفحة ولا يوضع تحتها خط وتكتب بحروف كبيرة عندما تكون بالانجليزية .

٩ـ يتبع الاسلوب الاتي عند كتابة المصادر على الصفحة الخاصة بالمصادر ترقم المصادر حسب تسلسل ورودها في البحث يكتب الاسم الاخير (اللقب) للباحث او الباحثين ثم مختصر الاسمين الاولين فعنوان البحث مختصر اسم المجلة المجلد او الحجم العدد الصفحات (السنة) وفي حالة كون المصدر كتابا يكتب بعد اسم المؤلف والمؤلفين عنوان الكتاب الطبعة . الصفحات . (السنة) الشركة الناشرة . مكان الطبع .

رقم الايداع في دار الكتب والوثائق ببغداد ٢٧٨ لسنة ١٩٩٠

البحوث العربية رقم الصفحة

السلوك الحياتي ليرقة طفيلي المشوكة الحبيبية *Echinococcus granulosus* من عترة الجمال في الفئران المثبطة مناعياً .

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(استلم بتاريخ ١٩٩٣/٩/١٥ ، قبل للنشر في ١٩٩٤/١/١٨)

ABSTRACT

The study concerned with the biological behaviour that include (number, size and location of secondary hydated cysts) of the larval stage of *Echinococcus granulosus* from camel origin in laboratory immunosuppressed mice by irradiation with (Cobalt - 60) or by splenectomy, and compared with control mice, after injection of protoscolices intraperitonealy. The number and size of secondary hydatid cysts increased with age of infection in immunsuppressed mice more than control mice. Irradiated mice had higher number and size of secondary hydatid cysts compared with splenectomized mice. It seemed from this study the irradiation with (cobalt-60) was more effective as immunosuppressor on mice than splenectomy.

الخلاصة

تضمنت الدراسة السلوك الحياتي الذي يشمل (اعداد ومواقع وحجوم الاكياس الثانوية) ليرقة طفيلي المشوكة الحبيبية *Echinococcus granulosus* من خلال حقن الرؤوس الاولية (Protoscolices) لعترة الجمال في التجويف الخلبي للفئران المثبطة مناعياً باشعة كاما (كوبلت-٦٠) او باستئصال الطحال ومقارنتها مع مجموعة السيطرة . ازداد عدد وحجم الاكياس الثانوية في مجاميع الفئران بتقدم عمر الخمج وكانت الزيادة في العدد والحجم اكثر في الفئران المثبطة مناعياً مقارنة مع مجموعة فئران السيطرة . كذلك فقد كانت الزيادة في الفئران المثبطة بأشعة كاما اكثر مما هي عليه في الفئران المستأصلة الطحال. يظهر من هذه الدراسة أن التعرض للأشعاع له تأثير تثبيطي على الفئران اكثر من استئصال الطحال وكان ذلك واضحاً في اعداد وحجوم الاكياس الثانوية المكتظة .

المقدمة

يعد مرض الاكياس الصدرية من الأمراض الواسعة الانتشار في العالم ومن الأمراض المتوطنة في العراق استناداً على الدراسات التي اجريت فيه (١، ٢، ٣، ٤، ٥،) ان مصطلحي داء المشوكات Echinococcosis وداء العداري Hydatidosis يستخدمان للتعبي عن الخمج المشترك بين الأنسان والحيوان والذي يتسبب عن الطور البالغ الوسطي metacestodes للديدان الشريطية جنس المشوكات Echinococcus التابع الى عائلة التينيا Taeniidae (٦) . كما يعد نوع المشوكات الحبيبية

Echinococcus granulosus المسبب الرئيسي للأكياس الصدرية في قطرنا (٣) وتشير بعض التقارير الى وجود الطور اليرقي للنوع Echinococcus multilocularis في المنطقة الشمالية في قطرنا في كبد امرأة (٧) .

تكمن الأهمية الأقتصادية للخمج بالخسائر المتسببة عن تلف الأعضاء الخمجة لكونها غير صالحة للأستهلاك البشري بالأضافة الى كونها مصدراً لخمج الكلاب (٨) .

وكذلك يؤدي الى تأثيرات على الحالة الصحية للحيوان مما يؤدي الى قلة في انتاج الحليب والصوف وبطئ النمو والخصوبة في الحيوانات الداجنة (٩) .

تتكون الأكياس الثانوية نتيجة انفجار الكيس وتحرر الرؤوس الاولية داخل الجسم مما يؤدي الى حدوث اكياس عدرية مشابهة للأصل . أما تكوين الأكياس العدرية الثانوية في الحيوانات المختبرية فيتم بواسطة حقن مكونات الكيس الخصب في التجويف الخلبي (١٠) .

أجريت بعض الدراسات عن تأثير المثبطات المناعية على نمو وتطور الأكياس الثانوية في الفئران والمتسببة عن يرقات طفيلي Echinococcus multilocularis (١١) . كما درس تأثير استئصال الطحال وتأثير اعطاء مركبات الكورتيزون على تطور الأكياس الثانوية المتسببة عن يرقات نفس الطفيلي (١٢) .

المواد وطرائق العمل

استعمل في هذا التجربة ٦٠ فأر مختبري ابيض باعمار واوزان تتراوح من ٢٠-٣٠ يوم ومن ١٨-٢٥ غم على التوالي، وقد قسمت الفئران الى ثلاث مجاميع متساوية مكونة من ٢٠ فأر. المجموعة الاولى ثبطت مناعيتها بتعريضها الى جرعة اشعة كاما مقدارها ١٥٠ راد ولمدة ١,٣٤٥ دقيقة وكانت مساحة حقل التشعيع ١١x١٤ سم[٢] ، اما المجموعة الثانية فقد ثبطت مناعية باستئصال الطحال ومجموعة ثالثة استعملت كفئرا سيطرة.

- حقنت فئران المجاميع الثلاث بجرعة ١ ملليليتر من سائل الاكياس المدرية المستخلص من الاعضاء الخمجة في الجمال والحاوي على (٢٠٠٠) راس في التجويف الخلبي.
- اجري قتل وتشريح (٦) فئران من مجاميع التجربة الثلاث (فارين من كل مجموعة) وعلى فترات متعاقبة (كل ١٠ ايام)، ابتداءا بعد العشرة ايام الاولى من الخمج ولغاية (١٠٠) يوم بعد الخمج لملاحظة نمو وتطور الاكياس الثانوية، بما في ذلك اعداد ومواقع وحجوم الاكياس الثانوية في الفئران المثبطة مناعيا وفئران السيطرة.
- استخدم اختبار تحليل التباين (Analysis of variance) لمقارنة اعداد الاكياس الثانوية في مجاميع الفئران الثلاث.

النتائج

شملت نتائج الدراسة التغيرات البايولوجية (نمو وتطور) يرقات طفيلي *E. granulosus* من عترة الجمال في الفئران المثبطة مناعيا بالاشعاع او استئصال الطحال ومقارنتها مع مجموعة السيطرة.

لوحظ الخمج في الفئران المثبطة بالاشعاع في اليوم ٢٠ بعد اعطاء الرؤوس الاولية، ولوحظ في الفئران المثبطة باستئصال الطحال وفي مجموعة السيطرة في اليوم ٣٠.

ازداد عدد وحجم الاكياس الثانوية بتقدم عمر الخمج، حيث كان اعلى عدد وحجم للاكياس الثانوية في اليوم (١٠٠) بع الخمج للمجاميع الثلاث. كذلك فان اعداد وحجوم الاكياس الثانوية في الفئران المثبطة

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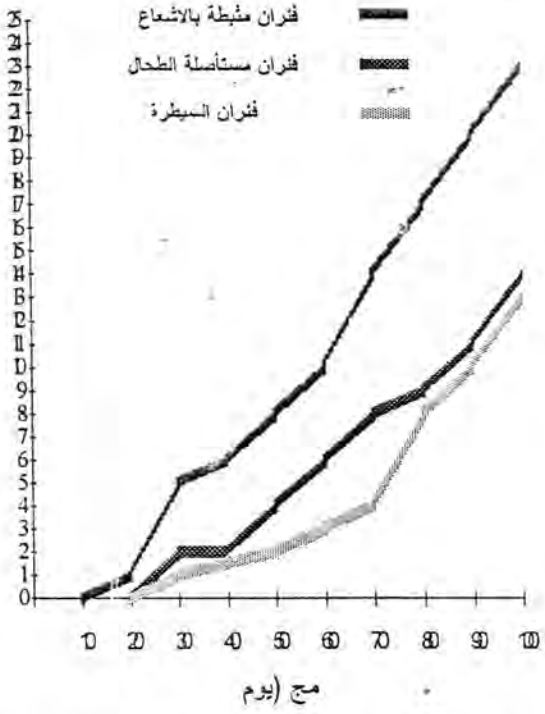
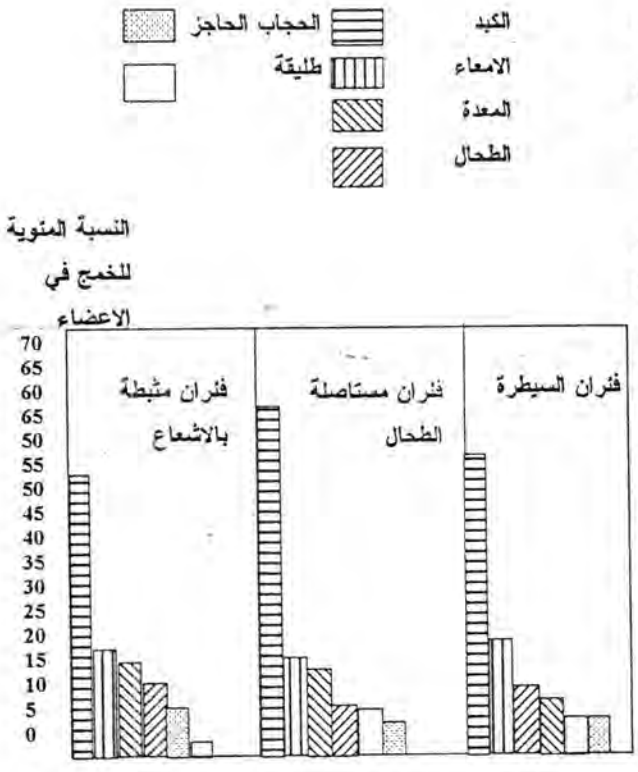
* اجري تشعيع فئران التجربة في معهد الاشعاع والطب الذري / بغداد

بالاشعاع اكثر مما هي عليه في الفئران المثبطة باستئصال الطحال، وكلا المجموعتين اعلى مما هي عليه في مجموعة السيطرة، وكما موضح في الجدول رقم ١.

بلغ اعلى معدل لعدد الاكياس الثانوية في المجموعة المثبطة بالاشعاع والمجموعة المستاصلة الطحال ١٣،٢٤ على التوالي في اليوم ١٠٠ بعد الخمج، والمجموعة المستاصلة الطحال ١٣،٢٤ على التوالي في اليوم ١٠٠ بعد الخمج، وبلغ في مجموعة السيطرة ١٢،٥ في نفس اليوم بعد الخمج، وهذا ما موضح في الشكل البياني رقم ١. وقد اظهر التحليل الاحصائي للنتائج وجود فروقات احصائية



شكل رقم (١) : معدل عدد الاكياس العددية الثانوية في الفئران المثبطة مناعياً بالاشعاع او بأستئصال الطحال وفئران السيطرة



مخطط بياني رقم ٢ : النسبة المئوية للخمجة في الاعضاء المختلفة في الفئران المثبطة مناعيا بالاشعاع او باستئصال الطحال وفئران السيطرة.

المناقشة

أظهرت نتائج هذا البحث زيادة في عدد وحجم الاكياس الثانوية في مجموعة الفئران المثبطة مقارنة مع مجموعة السيطرة . حيث بلغ اعلى معدل لعدد الاكياس الثانوية في الفئران المثبطة بالاشعاع ٢٤ كيس في اليوم ١٠٠ بعد الخمج ، في حين بلغ في فئران السيطرة ١٢،٥ كيس في نفس اليوم بعد الخمج . ان هذه النتائج جاءت مطابقة مع ما أشار اليه (١١) في ارتفاع نسب الخمج وانتشاره وزيادة معدل عدد الاكياس الثانوية في الفئران المعرضة للاشعاع والمخمجة تجريبياً بيرقات طفيلي *E. multilocularis* كما ان هذه النتائج جاءت مطابقة مع ما اشارت اليه (١٣) في الدراسة المجرات في العراق حول طفيلي *Hymenolepis nana* في ان مناعة الفئران تنحطم باستعمال التشعيع حيث ظهر جيلان من الديدان في

الفئران المعرضة للاشعاع في حين ظهر جيل واحد من الديدان في فئران السيطرة حيث ان اشعة كاما المستعملة ادت الى زيادة عدد البيوض التي كونت عدد كبير من الديدان .

اشارت نتائج البحث الى ارتفاع عدد وحجم الاكياس الثانوية في الفئران المثبطة مناعياً باستئصال الطحال مقارنة مع مجموعة السيطرة وان هذه النتائج جاءت مطابقة مع ما اشار اليه (١٤) في دراسته المجراة في العراق حول طفيلي *H. nana* حيث ازدادت نسبة الخمج بالطفيلي وازدادت اعداد الطفيلي في الفئران المستأصلة الطحال مقارنة مع فئران السيطرة حيث ان الطحال يلعب دوراً كبيراً في الحفاظ على المقاومة الطبيعية للجسم ضد بعض الامراض المتسببة عن الجراثيم والابتدائيات وحيدة الخلية وكذلك بعض الطفيليات متعددة الخلايا مثل ديدان الكبد وطفيليات *H. nana* (١٥) وجاءت غير مطابقة مع ما اشار اليه (١٢) في ان يرقات طفيلي *E. multilocularis* لايزداد نموها وتطورها في الفئران المستأصلة الطحال . ويمكن ان نستنتج من هذه الدراسة بان الاشعاع باشعة كاما له تأثير تثبيطي على الفئران اكثر من استئصال الطحال وهذا ما كان واضحاً في معدل عدد الاكياس الثانوية وحجمها الذي بلغ ٢٤ كيس و ٥-٦،٥ ملم على التوالي في الفئران المثبطة بالاشعاع في اليوم ١٠٠ بعد الخمج وبلغ ١٣ كيس و ٣-٤ ملم على التوالي في الفئران المستأصلة الطحال في نفس اليوم بعد الخمج ، حيث ان اشعة كاما تؤثر على خلايا اللمفوسايت في الأعضاء اللمفاوية وفي الدورة الدموية (١٦) ، وان التشعيع الموضعي بعنصر الكوبلت المشع يحدث تأثيرات كثيرة وحادة كانخفاض وهبوط المكونات الدموية ومنها اللمفوسايت والصفائح الدموية في الدم المحيطي، كذلك فان الاشعاع يثبط الخلايا القابلة للاستجابة المناعية والخلوية للجسم ويؤدي الى تأثيرات حادة على تكوين الدم ويؤثر على خلايا اللمفوسايت من نوع -ت-(١٧) .

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جدول رقم (١) : أعداد ومواقع وحجوم الاكياس العددية الثانوية في الفئران المثبطة مناعياً وفئران السيطرة .

نسل فار	وقت التشريح يوم بعد الخمج	نوع المعاملة	عدد الاكياس ومواقعها					مواقع اخرى	حجوم الاكياس (ملم)
			الكبد	المعدة	الامعاء	الطحال	طليقة		
١ ، ٢	١٠	(أ)	- ، -	- ، -	- ، -	- ، -	- ، -	- ، -	- ، -
		(ب)	- ، -	- ، -	- ، -	- ، -	- ، -	- ، -	- ، -
		(جـ)	- ، -	- ، -	- ، -	- ، -	- ، -	- ، -	- ، -
٣ ، ٤	٢٠	(أ)	- ، -	- ، -	- ، -	- ، -	٢ ، -	- ، -	٠،٥ ، -
		(ب)	- ، -	- ، -	- ، -	- ، -	- ، -	- ، -	- ، -
		(جـ)	- ، -	- ، -	- ، -	- ، -	- ، -	- ، -	- ، -
٥ ، ٦	٣٠	(أ)	٥ ، ٥	- ، -	- ، -	- ، -	- ، -	- ، ١٠ اح ح	٠،٥
		(ب)	- ، -	- ، -	- ، -	- ، -	٣ ، -	- ، -	- ، ٠،٥
		(جـ)	- ، -	- ، -	- ، -	- ، -	- ، -	- ، -	- ، ٠،٥
٧ ، ٨	٤٠	(أ)	٦ ، ٧	- ، -	- ، -	- ، -	- ، -	اح ح ، -	٠،٥ - ١
		(ب)	١ ، ١	- ، -	- ، -	- ، -	٢ ، -	- ، -	٠،٥ - ١،٥ ، ٠
		(جـ)	- ، ١	- ، -	- ، -	- ، -	١ ، ١	- ، -	٠ ، ٥
٩ ،	٥٠	(أ)	٥ ، ٧	- ، ١	٣ ، ٢	٢ ، -	- ، -	- ، -	١ - ١،٥
١٠		(ب)	٢ ، ٣	- ، ٢	- ، -	- ، -	- ، -	- ، -	١ - ١،٥
		(جـ)	١ ، ٢	- ، -	- ، -	- ، -	١ ، -	- ، -	١ ، ٥ - ١
١١،١	٦٠	(أ)	٥ ، ٨	٣ ، ٢	٣ ، -	١ ، ١	- ، -	- ، -	٢ - ٢،٥
٢		(ب)	٤ ، ٢	- ، ٢	٢ ، -	- ، -	- ، -	اح ح ، ١٠	١،٥ - ٢،٢
		(جـ)	٢ ، ٣	- ، -	١ ، -	- ، -	- ، -	- ، -	١ - ١،٥
١٣،١	٧٠	(أ)	٦ ، ٧	٢ ، -	٤ ، ٣	٣ ، ٢	- ، -	٣ح ح ، ١٠	٣ - ٢،٥
٤		(ب)	٣ ، ٤	- ، -	٣ ، -	- ، -	- ، -	اح ح ، -	٢ - ٢،٥
		(جـ)	٣ ، ٣	١ ، -	- ، -	١ ، ٢	- ، -	- ١٠ اح ح	٢-١،٢-١،٥
١٥،١	٨٠	(أ)	٥ ، ١١	٥ ، ٢	٥ ، ٣	٢ ، ١	- ، -	- ٢٠ح ح	٤ - ٣
٦		(ب)	٤ ، ٦	٣ ، ١	- ، -	- ، -	- ، -	- ، -	٣ - ٢،٥
		(جـ)	٣ ، ٤	١ ، -	١ ، ٢	١ ، ٢	- ، -	- ، -	٢ - ٢،٥
١٧،١	٩٠	(أ)	١٠ ، ١٢	٤ ، ٢	٥ ، ٣	٤ ، ٢	- ، -	- ، -	٥ - ٤،٥
٨		(ب)	٥ ، ٦	٢ ، ٢	٢ ، ١	- ، -	- ، -	اح ح ، -	٣ - ٣،٥
		(جـ)	٣ ، ٨	- ، -	٣ ، ١	١ ، ٢	- ، -	اح ج ، -	٢-٣ ، ٥،٣-٣
١٩،٢	١٠٠	(أ)	١٠ ، ١٢	٥ ، ٣	٨ ، ٢	٣ ، ٣	- ، -	٢ح ح ، -	٥ - ٦،٥
٠		(ب)	٨ ، ٨	١ ، ٢	٣ ، ٣	- ، -	- ، -	اح ح ، -	٣ - ٤
		(جـ)	٩ ، ٣	٣ ، ١	٤ ، ٢	٢ ، -	- ، -	- ١٠ اح ج	٢-٥ ، ٣،٥-٢، ٣،٥

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تحضير مستخلص M2 لاستخدامه في تشخيص الارجية الناجمة عن بعض انواع الفطريات

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ABSTRACT

The sim of this study was to prepare allergenic extract, from some species of fungi included in M_2 extract using extraction-solution, which can be used in skin test for identification of fungal allergy. The potency of the extract was tested, an intraperitonial sensitizing dose followed by intratrachial shocking dose were injected in 7 males (300-350 gms) Guinia-pigs. The results showed a significant increase in the absolute number of eosinophils in periphral blood compared with the normal control. Histopathological studies revealed an infittration of traches with eosinophils and neutrophils leukocytes .

الخلاصة

الهدف من الدراسة هو تحضير مستخلص ارجي من بعض انواع الفطريات التي تتضمنها خلاصة M2 باستخدام محلول استخلاص ، يمكن استخدام هذا المستخلص في اختبارات الجلد لتشخيص ارجية الفطريات . تم اختبار كفاءة هذا المستخلص بحقن جرعة محسسة داخل البريتون وجرعة صادمة داخل القصبة الهوائية باستخدام 7 من خنازير غنيا ذكور بوزن ٣٠٠-٣٥٠ غم . اوضحت نتائج الاختبار زيادة معنوية في عدد خلايا الدم الحمضة في حيوانات الاختبار مقارنة بحيوانات السيطرة . ظهر من الدراسة النسيجية للقصبة الهوائية ارتشاح واضح للخلايا الحمضة والعدلة .

المقدمة

هناك قائمة من المستأرجات المحمولة في الهواء (Air born allergens) من امثلتها حبوب الطلع ، سبورات الفطريات ، حلم غبار المنزل (House dust mits) شعر الحيوانات وقشورها ، ريش الطيور .

وان لمثل هذه المستأرجات القدرة على حث الجسم على انتاج اضداد نوع IgE ذلك لدى الاشخاص المحسسين لها (Allergic Peoples) وهناك عدة عوامل تلعب دوراً مهماً في احداث الارجية منها :

أ- وفرة هذه المستأرجات في البيئة ، حيث لايكفي كون المريض ذو استعداد لاظهار تفاعل الارجية بل يجب ان تكون المستأرجات التي تثير التفاعلات الارجية لديه موجودة وبكمية كافية لتكون بتماس مع المريض .

ب- حجم المستأرجات وهذا بالنسبة لارجية الجهاز التنفسي حيث ان اغلب المواد المحمولة في الهواء تتراوح اقطارها بين 2-60 مايكرون ، فان ملائمة حجم الجزيئات مهم للوصول الى الجهاز التنفسي .

جـ- طبيعة المستأرج ، تحتوي هذه المواد مكونات تركيبية عديدة فقط قسم قليل منها تكون مستضدية

والتي هي عادة مكونات بروتينية مع قليل من السكريات، وذات وزن جزيئي يتراوح بين 40000-3000 دالتن .

من هذه المستأرجات ما تستطيع بمفردها حث الجسم على انتاج الـ IgE وتسمى هذه مستضدات كاملة البروتين (Complete Pretein antigems) والتي من امثلتها الفطريات ، والتي يمكن استخدامها في اختبار الجلد للتحري عن الارجية ، حيث ان ارتباط الـ IgE بالخلايا المستهدفة يتيح لنا امكانية الكشف عن الارجية وذلك في حالة كون هذه الاضداد هي نوعية للمتأرج موضوع البحث .

طرق اختبار الارجية

١- *اختبار الجلد Skintest*

٢- *اختبارات التحدى (المحرض)*

Challenge (Provocation)

أ- تحدى الانف Nasal Challeng

ب- التحدى القصبي Bronchial Challenge

٣- *عد الخلايا الحمضة Eosinophils counting*

اختبار تحدى الاستنشاق

(Inhalation challenge test)

سجل بلاكلي Blackly .. (1) قديما استجابة تقلي قصبي عند لكلا من الرشاشي Penicillium والـ Chetomium .

ان تعريض العضو المستهدف مباشرة بالمستأرج يعد ذو اهمية في اثبات ارجية المادة المراد اختبارها .

فلقد سجل هاريز Harries .. (2) نتائج تجدى استنشاق اجراه على 22 مريض باستخدام ابواغ الـ Alternaris

وسجل بنكتون Penington (3) نتائج تعريض موجبة في مجموعة من 526 مريض بتعريضهم لمسحة محملة بالابواغ داخل الانف وكذلك تطبيق مباشر لخلاصة المستأرج داخل الانف .

يظهر التفاعل الموجب على شكل تغير في الاغشية المخاطية للجهاز التنفسي وتردد العطاس وحك الانف وظهور افرازات من الانف وصعوبة في التنفس ، وان المادة المستخدمة في التحدى يمكن ادخالها بشكلها الطبيعي او تستخدم كخلاصة تحضر بطريقة مناسبة من المستأرج الطبيعي بعد ان يتم تقبيسها .

اما اوكودا Okuda .. (3) فانه يفضل تطبيق قرص ورقي مشبع بالمستأرج ويترك حتى يجف والذي يكون به محتوى معلوم من المستضد 250 مايكروغرام .

ان ايصال كمية من المستأرج مباشرة الى القصبة الهوائية يعطي معلومات مباشرة عن اهمية المستأرج النوعي وان الاستجابة للاختبار تعكس مدى تحسس المريض ويستخدم بشكل رئيسي للاغراض البحثية ، اما سريرياً فهناك تحديدات في الموضوع بسبب تردى اعراض الربو عند استخدامه ويجرى في المراكز المتخصصة وتحت اشراف خبراء .

بعد استنشاق المستأرج بـ 5-10 دقائق تظهر الاعراض ويصل اقصاها بعد 20-30 دقيقة .

وقت ظهور الاعراض قد تشابه تفاعل الجلد الفوري لكن الفسيولوجيا المرضية تختلف عما في الجلد وقد يظهر تفاعل متأخر بعد 4-8 ساعات من الحث بالمستأرج وقد يظهر تفاعل ارثس .

ان تحضير خلاصة خالية من السمية ومعقمة من الفطريات وغيرها من الارجينات محلياً واثبات كفاءتها لاستخدامها في اختبارات الجلد في مرض الارجية لتحديد أي المستأرجات وراء تحسيسهم ، يحمل اهمية بالغة حيث يمكن ان تحل هذه الخلاصات محل الخلاصات المستوردة ولما يمر به القطر من حصار جائر ، جاءت هذه المحاولة وهي الاولى من نوعها في العراق متوافقة مع توجه البلد نحو تحقيق الاكتفاء الذاتي .

المواد وطرق العمل

١- انواع الفطريات التالية *:

Phizopus stolonifer
Stemphylium consortiale
Rhizopus nigricans
Mucor racemosus
Mucor spinsus
Botrytis cimerea

٢- جرى تنمية الفطريات المشار اليها في وسط السابورد السائل Sabouraud's broth في دوارق Thompson وبدرجة حرارة 25 م° لمدة 3 اسابيع ثم سكب الوسط الغذائي ، ورطبت الحصيرة الفطرية بكحول مثيلي وجففت الكتلة النامية بدرجة حرارة 56 م° وبعدها تم سحقها باستخدام مجرشة القهوة.

٣- محلول الاستخلاص : الطريقة معتمدة في مختبرات انتاج اللقاحات في معهد باستور والمشار اليها في (5)

NaH_2PO_4	0.37 gm.
Na_2HPO_4	1.42 gm.
NaCl	2.5 gm.
Phenol	4.0 gm.
D.W.	1 liter

حضر 10٪ حجم من الفطريات مع محلول الاستخلاص وترك بدرجة حرارة 4 م° لمدة اسبوع مع التحريك بين حين وآخر ، بعدها تم اجراء طرد مركزي للمستخلص 3600 دورة لمدة 20 دقيقة ثم رشح العالق واجريت للراشح عملية ديال (Dialysis) لمدة 48 ساعة ضد ماء لا ايوني (Deionized water) ثم جرى تحديد التركيز وزن / حجم حسب الطريقة التي اشار اليها Alam (6)

استخدمت طريقة التجفيد (Lyophilization) اثناء اعداد الخلاصة .

تم ضبط الاس الهيدروجيني للخلاصة الى 7.4 ، عقمت الخلاصة بمرشحات فائقة 0.22 مايكرون.

جرى التاكد من كون الخلاصة عقيمة وخالية من السمية باتباع الطريقة المشار اليها في (7) .

الاختبارات في الحيوانات التجريبية

تم اجراء الاختبارات على 7 من خنازير غينيا ذكور بوزن 300-350 غم (تم الحصول عليها من مختبر الرقابة الدوائية في وزارة الصحة) وكانت وفق التسلسل التالي:-

١- حساب اعداد خلايا الدم الحمضة ، استخدمت طريقة Dacie and Lewis (8).

٢- تحسيس الحيوانات بحقن حجم 0.2 سم^3 من الخلاصة بتركيز وزن / حجم داخل الصفاق .

٣- الجرعة الصادمة (Shocking dose) بعد مضي 14 يوم على تحسيس الحيوانات تم حقنها بـ 0.01 سم^3 من الخلاصة بتركيز 1000:1 وزن / حجم داخل الرغامي .

٤- تمت مراقبة الحيوانات وسجلت العلامات المرضية وبعد مضي 15 دقيقة تم سحب ١ سم^3 من الدم من قلب الحيوانات لحساب اعداد الخلايا الحمضة . وبعد 20 دقيقة اخذت قطع من القصبة الهوائية لقسم من الحيوانات والقسم الاخر بعد 6 ساعات لاجراء الدراسة النسيجية (9) .

النتائج

ظهر من اختبار المقاسة كون الخلاصة عقيمة حيث لم يظهر اى نمو على اوساط الـ (T.C.A) الهوائية واللاهوائية اما بخصوص السمية فظهر انها غير سامة .

تم تسجيل علامات مرضية عديدة بدت على الحيوانات التي جرى تعريضها الى الجرعة الصادمة من الخلاصة وكانت هذه العلامات المرضية متماثلة في الشدة وهي :-

١- *صعوبة وعدم انتظام التنفس*
٢- *زيادة ضربات القلب*
٣- *سعال متكرر*
٤- *نفش الشعر وانزواء الحيوان*
٥- *تبول متكرر*

* تم عزل انواع الفطريات اعلاه من الدكتور علي البهادلي-كلية الزراعة-جامعة بغداد .

وقد رافق هذه العلامات زيادة واضحة وبفوارق معنوية في اعداد خلايا الدم الحمضة حيث كان متوسط عددها قبل اجراء الاختبار 430 ±14.71 خلية لكل ١ سم^{3} من الدم وارتفع متوسط عددها الى 3053 ±85.23 خلية لكل ١سم^{3} من الدم ويتضح ذلك بالتفصيل ولكل حيوان من الجدول رقم (1) .

اوضح التحليل الاحصائي باستخدام اختبار T)- (test احادى الطرف وجود فرق معنوي بين حالتي المقارنة (قبل وبعد المعاملة) عند مستوى احتمالية اقل من 0.005 وخمسة درجات حرية .

جدول ١ يبين اعداد الخلايا الحمضة لكل ١ سم^{3} من الدم قبل وبعد المعاملة بالمستخلص

رقم الحيوان	عدد الخلايا الحمضة قبل المعاملة	الانحراف القياسي	عدد الخلايا الحمضة بعد المعاملة	الانحراف القياسي
1	435		2905	
2	405		3035	
3	440		3080	
4	415		3090	
5	440		3030	
6	440		3190	
7	430		3040	
المتوسط	430	±14.71	3053	±85.23

شكل (١) مقطع نسيجي للرغامي في خنزير غينيا حضر المقطع بعد -٦- ساعات من المعاملة بالجرعة الصادمة.

شكل (٢) مقطع نسيجي للرغامي في خنزير غينيا طبيعي صبغ المقطع بالايوسين والهيماتوكسلين.

الدراسة النسيجية

ظهر من دراسة المقاطع النسيجية المحضرة من الرغامي للحيوانات التي اجرى عليها الاختبار ارتشاح واضح لخلايا الدم الحمضة وُالخلايا العدلة وذلك بعد 20 دقيقة من التعريض للجرعة الصادمة حيث اشير بالعلامة (+) لخلايا الدم الحمضة وبالعلامة (±) للعدلة واصبح هذا الارتشاح اكثر في المقاطع المحضرة بعد 6 ساعات من التعريض للجرعة الصادمة ، وأشير بالعلامة (+ +) بالنسبة لخلايا الدم الحمضة والعلامة (+) بالنسبة للعدلة وكما في الشكل (1) في حين لم يظهر اى ارتشاح في المقاطع المحضرة لحيوانات السيطرة وكما في الشكل (2) .

المناقشة

تبين من خلال اجراء التحليل الاحصائي لنتائج اختبارات الجلد لـ 1200 مريض وعلى مدى 12 شهر والتي يتم اجراءها في المركز التخصصي لامراض الحساسية والربو ان نسبة التحسس لخلاصة M_2 حوالي 4.2& وذلك من بين حالات التحسس الموجبة ورغم قلة نسب المرضى المتحسسين للفطريات الداخلة في تركيب خلاصة M_2 والتي هي قليلة الشيوع في البيئة الا ان اهميتها تكمن في حصول التعرض لها

موضعياً حيث في ظروف معينة يحصل التعرض لكميات هائلة من ابواغ هذه الفطريات. وجاءت هذه المحاولة بعد ان تم تحضير خلاصات اهم المستأرجات الشائعة في البيئة العراقية (7) .

تم التاكيد على المستأرجات المحولة في الهواء وذلك لعدم امكانية تجنبها مقارنة مع المستأرجات الاخرى حيث يتم التعرض لها عن طريق الاستنشاق .

جاء الثناء على الخلاصات المحضرة محليا من قبل عدة باحثين فلقد ذكر ساندو Sandhu (10) انه قام بمقارنة خلاصة حضرها من الفطريات محلياً وقارنها مع خلاصة مستوردة واجرى لها اختبار على 240 مريض فوجد ان 32% من المرضى اسجابوا للخلاصة المحلية مقابل 30% استجابوا للخلاصة المستوردة وهي اوسع فائدة سواء من ناحية اختبارات الجلد او العلاج المناعي ، يفضل العديد من الباحثين استخدام خلاصة المستأرج في تاكيد ارجية المادة المراد دراستها (11) .

جاء التاكيد على دور الخلايا الحمضة في الارجية وعلاقتها الوثيقة مع اعراض الربو (Asthma) (وذلم بسبب احتواءها على العديد من الوسائط) (12) كذلك الحال بالنسبة لخلايا الدم العدلة حيث ذكر لارسون Larson (13) ان هذه الخلايا لها دور في الربو وخصوصا في الطور المتأخر .

وبذلك يمكن اللجوء في الاستدلال على شدة الاستجابة للمستأرج بمتابعة ارتشاح الخلايا الالتهابية بالاخص الخلايا الحمضة والعدلة سواء كان ذلك في الدم المحيطي او من خلال متابعة ارتشاحها في المقاطع النسيجية حيث ان الخلايا الحمضة تقضي وقتا في الدم خلال رحلتها من نقي العظم الى مواقع التفاعل في الانسجة استجابة لعامل الجذب الكيمياوي لها eosinophils chemotaotic factor وكذلك الحال بالنسبة للخلايا العدلة التي تتأثر بعامل الجذب الخاص بها Neutrophils chemotactic factor اضافة الى امكانية متابعة الاعراض المرضية الناجمة عن المعاملة بالمستأرج .

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دراسة تصنيفية للجنس Asperuge L. في العراق

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ABSTRACT

The present study of the Monotypic Asperugo L. coners all its attributes and detailed morphology. The plants of this genus are mostly creeping weeds with quadrangular flaccid stems, and covered with strigose and hispid hairs. Out of the characteristics are the shape of flowering and fruiting calyxes which are compressed and with small secondary lobes between the main calyx lobes, presence of faucsl appendages, pyriform nutlets and the way it connected with fruiting stalk. Pollen are often irregular, faintly trisulcate, protandry and crossing are dominant. These plants prefer growing in a dump shaded places near water streames and fields and orchard with surplus of irregation.

الخلاصة

تغطي الدراسة الحالية للجنس Asperuge L. الوحيد النوع ، خواص نباتات هذا الجنس ونوعه ومظاهره الخارجية وبشكل دقيق ومفصل ، فقد وجد ان نباتاته غالبا زاحفه وذات سيقان مضلعه رخوه وسهنه التكسر ويغطي سطح النبات شعيرات شوكية قاسيه . ومن مميزات النوع البارزه هي شكل الكأس الزهري والكأس المضغوط جانبيا والذي توجد بين فصوصه الرئيسه فصوصا ثانوية صغيرة كذلك يمتاز بوجود الزوائد اللهاتيه ، وبشكل البندقيات الثمرية الكمثرية وطريقة اتصالها بالحامل الثمري ، الشكل غير المنتظم لحبوب لقاحه الثلاثية الاخاديد غير الواضحة وبوجود ظاهرة الذكوره المبكرة Pretandry لازهاره وسياده التلقيح الخلطي فيها . وان نباتاته تفضل في نموها الترب الرطبة والمظللة وترب مجاري المياه وعلى حافات البساتين والمزارع غزيرة الري .

المقدمة

يضم الجنس Asperuge L. من العائلة (Beraginaceae) حسب ما جاء في لينيوس (1) Linnaeus نوعا واحدا هو Precumbens L. تنمو نباتاته في اوربا . وأعطى بنثام وهوكر (2) Bentham & Hooker وصفا موجزا جدا للجنس فقط مشيرا الى نوعه الوحيد النامي في اوربا وآسيا . وكتب بواسيه Boissier (3) ان الجنس نوعا واحدا وضربا هو *A. precumbense var. peduncularis* حيث جمعه بواسيه من بلاد فارس واشار الى ان هذا الجنس ينمو في مناطق مُن اوربا وسيبريا وشمال افريقيا ، وقد اورده بوست Post (4) ضمن نباتات سوريا وفلسطين وسيناء . وكتب هاتسن Hanson (5) وصفا مقتضيا لهذا الجنس ونوعه ضمن النباتات النامية في اوربا موضحا الدول الاوربية التي بنيت فيها ، ثم ذكره ويليس Willis (6) في قاموسه كجنس ذو نوع واحد تنمو نباتاته في اوربا . وحسب ما ورد في ريدل Riedl (7) وادموندسن Edmondson (8) فان نفس النوع A. precumbens ينمو في الاراضي الايرانية والاراضي التركية على التوالي ، وقد اشار كريم

وقرعان (9) في ازهار الاردن البريه الى هذا النوع ذاكرين مواقع وجوده في هذا البلد .

يظهر ان نباتات هذا النوع ، كما اشار كيت Guest (10) الى انها ذات ازهار زرق وتنتشر بكثرة في الاماكن البارده الرطبه المظلله وتحت الاشجار والجدران وتمتد في انتشارها الى مناطق تصل الى ارتفاع 1500 متر وتزهر خلال شهري آذار ونيسان وترعى هذه النباتات من قبل المواشي . وقد اشار الراوي (11) الى بعض مناطق نموه في العراق ، واقتصر رشنكر Rechinger (12) على وصف مقتضب للجنس ونوعه ذاكرا وجوده في بغداد وبابل فقط . اما جاكرافارتي Chakraverty (13) فقد اوجز عده اسطر عن مظاهر النوع الخارجية ذاكرا انه اعشاب رعوية وتفضل كعلف عندما توجد بكثره، وهي منتشره بغير نظام Straggling herbs في اوربا والشرق الاوسط وشبه الجزيرة العربية وشمال آسيا .

يتبين مما ورد آنفا انه لم يتطرق أي من المصادر التي حصلنا عليها الى اية تفصيلات حول مظاهره الخارجية مطلقا فضلا عن طبيعته وتوزيعه المتكامل في العراق ، لذلك فمن دواعي البحث اننا قمنا بدراسة هذا الجنس مظهريا وتوزيعيا مع اعطاء معلومات عن حبوب طلعه . ومما يدفع الباحثين الحاليين على الدراسة هو تضارب بعض الصفات الوارده لنباتات هذا الجنس في مختلف المصادر رغم تمييزها مظهريا .

المواد وطرائق العمل

اعتمدت الدراسة الحالية على النماذج المعشبيه المودعه في المعاشب العراقية وبالاخص معشب الجامعة (BUM) والمعشب الوطني العراقي (BAG) ومعشب كلية الزراعة في أبي غريب (BUA) ومعشب متحف التاريخ الطبيعي (BUN) وقد كان عدد النماذج التي درست 160 عينة حيث تم عزل وتشخيص جميع العينات المودعه فيها . وقد درست 20 عينة نباتيه طريه جمعت من جبال مقلوب وسنجار خلال سفره علميه تمت خلال نيسان 1993، واعتمدت هذه النماذج الطريه ايضا في قياس مختلف الاعضاء والاجزاء النباتية ثم حفظت في معشب الجامعة .

درست المظاهر الخارجية باستعمال مجهر التشريح الالماني Schotl حيث تم قياس ابعاد الاعضاء النباتية واجزائها ، اما الاجزاء النباتية الصغيرة جدا والدقيقة كالبندقيات والشعيرات وحبوب اللقاح فقد تمت دراستها باستعمال مجهر التشريح والمجهر الضوئي المركب Olympus الياباني . درست خصائص حبوب اللقاح بعد استخراجها من المتوك على شرائح زجاجية باستخدام طريقة السفرانين جلي كليسرين ، المياح (14) . استعملت خارطة العراق ذات المقاطعات الجغرافية والتي اوردها كيست Guest (15) لغرض توزيع نباتات هذا النوع على مختلف مقاطعات العراق الجغرافية ، واعدت رسوم تخطيطية لمختلف الاعضاء النباتية .

النتائج والمناقشة

١- الدراسة المظهرية وحبوب اللقاح :

من خلال مراجعة بعض المصادر يبدو ان اقرب اجناس العائلة Beraginaceae للجنس Asperuge هي Rochelia Reichb. و Omphalodes Moench و Myesetis L. و Amsinekia Lehm. حيث يرد عادة بين هذه الاجناس في معظم الموسوعات النباتية لكنه يتميز وبكل وضوح عن هذه الاجناس وغيرها بطبيعة الكأس الزهري والكأس الثمري في افراده وكذلك طبيعة النوره والسيقان المتفرعة الهشة الرباعية الاضلاع .

لافراد نباتات A. precunbens جذورا وتديه بنية اللون ذات طبيعة خشبية رغم انها حوليه ويتراوح طول الفرد النباتي الواحد 30-55 سم عادة وقد يتعداه ليصل المتر الواحد عند توفر البيئة الملائمة ويتفرع

الساق العشبي الغض والاخضر اللون والسريع التكسر Flaccid والمجوف والرباعي المقطع Quadrangular ، تفرعا منتشرا Diffusely branched عند القاعدة بحيث تبدو عدة سيقان تستلقي على الارض Precumbent وتتسلق قسما منها متى ما وجدت سندا لذلك ، ويتفرع كل فرع رئيسي للساق تفرعات ثانوية في أقسامها العليا . ويتراوح قطر الساق الذي يكون عاده متناسق 3-5 ملم ويغطي سطحه شعيرات شوكية Setae معقوفة عكس اتجاه نموه وللشعيرة الواحدة قاعدة غريضة شبه درنيه تتغاير اشكالها حسب موقعها على الساق وهي ذات لون ابيض ، واللون الابيض لقاعدة الشوكه يكون متغير فقد يكون داكن عند تمام النضج للنبات ، ويكون كريمي اللون عند بداية تكون قاعدة الشوكة . ولايتجاوز طول الشعيره الواحده 1 ملم وتبدو بشكل واضح على طول الاضلع الساقية . يبدو ان تفرع الساق بهذا الشكل ووجود الشعيرات القاسية وبهذه الطبيعة عوامل تساعد النبات على تثبيت موقعه على الارض او بين النباتات او على تسلقه مع فروعه على نباتات اخرى او ما شابه من المساند المجاورة .

تكون الاوراق متبادلة في الاجزاء السفلى والوسطى من السيقان ، اما في العليا وبداية تفرعاته فتكون متقابلة من آباطها براعم زهرية عادة . ويخلو الفرد النباتي من الاوراق القاعدية اما الاوراق الساقية Caulines فتميز العليا منها بكونها جالسة وخصوصا تلك التي تخرج من اباطها الازهار ، ويمتد نصل الورقة واحدة من الاوراق الساقية السفلى الى مسافة طويلة وضيقة للاسفل حيث يظهر سويق الورقة مجنسا ، ابعاد النصل 8-12 x 2.5-3 سم وطول السويق 2-3.5 سم ، اما ابعاد النصل في الاوراق العليا 2.5-6 x 1-2.4 سم وللنصل شكلا اهليليجيا او اهليليجي الى رمحي مقلوب Elliptic or Elliptic-oblanceolate وقمة النصل حادة Acute الى مقورة Obtuse وبعض القمم في بعض الافراد قمية قصيرة Shortly apiculate وقاعدة النصل ممتدة لذلك يبدو سويق الورقة مجنحا وحافة النصل مستقيمة Entire ، الشكل (1 – أ) .

تكون شعيرات الورقة قصيرة متوسطة الى قليلة الكثافة وتنعدم عادة على العروق عند سطح الورقة العلوي ، اما على السطح السفلي فتكون اقل كثافة وتستطيل على منطقة العروق . تكون الشعيرات على نوعين ، النوع الاول تخرج خلية الشعيرة من درنة Tubercle ملساء او مفردة لذا فهي Haplotrichous ، وتكون الدرنة ذات صف او صفين من الخلايا ، اما النوع الثاني فلا تخرج خلية الشعيرة من درنة وهي اقصر من النوع الاول ، وتكون قواعد الخلايا لشعيرية منتفخة رأسية الشكل Capitate ، الشكل (1 – ب) وبصورة عامة تكون الشعيرات قائمة Patent ذات لون ابيض لماع ، اما العروق فهي شبكية وبارزة على سطح النصل السفلي ومنخفضة على السطح العلوي .

تعد الاوراق الساقية العليا التي تخرج من آباطها الازهار قنابات ورقية Leafy bracts وتكون الازهار مفردة او ثنائية في ابط الورقة الواحدة Axillary وقد تخرج ثلاث ازهار من أبط ورقة واحدة خصوصا عندما يكون الفرد النباتي نشط في نموه ، وتبدو الشعيرات البيض في هذه الاجزاء المزهرة من النبات اكثف مما في بقية اجزاء النبات مما يزيد قابلية النبات على التعلق .

يتراوح طول حامل الزهرة بين 2-2.5 ملم وهو ذو شعيرات شوكية قصيرة تتجه للاعلى باتجاه الكأس الزهري وهي شبه منبطحة Subadpress . وللكأس خمسة فصوص وتكون نسبة تشققه 3/5 طوله ، ويتراوح طول الفص 2-3 ملم وعرضه 0.7-1 ملم وذو شكل رمحي وقمته حاده الى محتدة Acute to Acuminate وحافته مشعره خشنه Hispid. ويوجد بين الفصوص الرئيسه الخمسة للكأس فصوصاً ثانوية صغيرة ، الشكل (2-1). ويبدو الكأس قاربي الشكل مضموم ومفصص الحافات وذو شعيرات خشنه ايضا ، وقليل بينها من نوع Strigese وهي كثيفة بيض اللون

، ان شكل الكأس هذا يوضح وبشكل جلي وظيفته في المحافظة على الزهره فضلا عن القيام بالبناء الضوئي ، تسقط احيانا شعيرات الكأس في مرحلة الكأس الثمري فيبدو الاخير شبه ألمس Glabrescent. وتتميز للكأس شبكة واضحة جداً من العروق وخصوصاً عند مرحلة نضج الزهره وتنمو وتتوضح اكثر في مرحلة الكأس الثمري المتوسع .

يتميز التويج بشكله الجرسي Campanulate والذي يميل احيانا الى الشكل الجرسي الاسطواني Cylindriee-campanulate ، وهو أزرق عند نصفه العلوي وأبيض في نصفه السفلي والفاصل بين اللونين تخصر التويج ذو اللون البنفسجي الواضح او النيلي وهذا في الاشكال العراقية لهذا النوع ، وللنوع اصناف (اشكال) عديدة تختلف الوانها فهناك الصنف الشائع في السليمانية ذو الجزء الاعلى من التويج الوردي اللون الى البرتقالي والجزء السفلي بنفسجي ، وقد وصف ايدموندسون (8) Edmondson.

في الموسوعة النباتية التركية ، التويج بانه قمعي الشكل في الوقت الذي يظهر وبكل وضوح شكله الجرسي هذا ولم يشر هذا المصدر الى وجود هذا النوع في العراق ، ويصف هانسون Hanson (5) التويج بالقمعي ايضا في الموسوعة النباتية الاوربية وكذلك Boissier (3) و Post (4) اذ يبدو ان هؤلاء قد نقل بعضهم عن بعض دون التدقيق في فحص النماذج ، وربما يكون السبب هو تمييز الجنس ونوعه بحيث انه لايحتاج الى هذه الدقة في التشخيص او لدقة الازهار وعدم سهولة وصف اجزائها ، هذا ولم يشر (7) Riedl اصلا الى شكل التويج ، وتتراوح ابعاد التويج بين 3-2.5 x 1-0.75 ملم وهو خالي من الشعيرات كليا الا انه يحتوي على خمس زوائد لهاتيه Faucal appendages مهدبة ومثلثة الشكل تقريبا مع فصوصه البيضوية وتتبادل مع متوك الاسدية الواقعة تحت مستواها . ان معدل بروز التويج عن الكأس الزهري حوالي 1/2 ، وان الانبوب التويجي يكون اسطواني الشكل وخصوصا عند نصفه السفلي وهناك عند قاعدته والى الداخل عشر قطع كل منها مستطيلة الشكل ملساء وتمثل بمجموعها حلقة غدد الرحيق ، الشكل (2-5) .

يبدو ان ازهار هذه النباتات خلطية التلقيح Cressing بالرغم من عدم بروز الاسدية والمتوك خارج التويجات ، لان الاعضاء الجنسية متباينة النضج Dichogamy حيث ان المتوك تنضج تاثره حبوب طلعها قبل ان تكون المياسم مستعدة لاستلامها في نفس الزهره أي ان الازهار مبكرة الذكوره Pretandry . ويستدل ايضا على ان هذه الازهار تفضل التلقيح الخلطي بسبب وجود غدد الرحيق وتلون التويج وهاتين الصفتين من صفات الازهار الحشرية التلقيح ، وقد يحدث التلقيح الذاتي Selfing ، ورغم ذلك فأن هذه الظاهره تحتاج الى دراسة وتعمق اكثر للوقوف على حقيقتها وعلاقة ذلك بتطور نوع هذا الجنس وانفراده .

تظهر الاسدية المتساوية الطول وكأنها خمسة متوك فقط ملتصقه في الثلث الداخلي الاسفل من التويج تحت او عند منطقة التخصر وتحت اللهاتيه ، ذات ١ ملم طولا ، واسطوانية الى اهليليجية الشكل شبه جالسه وطبعا ستكون داخل التويج كليا ويتصل المتك الواحد الاصفر اللون بالخويط القصير جدا من منطقة ظهرية ويتفتح طولياً ويمكن الحصول على الخويط بصعوبة حيث يبدو خيطي الطبيعة املس لايتجاوز طوله 0.4-0.2 ملم ، الشكل (2-5) ، وتتفتح طولياً حيث تنثر الطلع .

تبدو حبوب الطلع تحت المجهر الضوئي ثلاثية الفصوص Trilobed وثلاثية الاخاديد Trisulcate ابعادها 10-7(8.97) مايكروميتر في المحور القطبي ، ومتطاوله Oblong او شبه كمثرية Subpyriferm او متطاولة مخصره Constricted elengate في المحور الاستوائي وابعادها 13-9(11.66) مايكروميتر ، أي ان حبوب الطلع هي من النوع الصغير جداً او الصغير ، اردتمان Erdtman (16) . ان حبوب طلع هذا النوع متغايرة الاقطاب Heteropelar وذات اخاديد

غير متميزة بوضوح ولايبدو تحت المجهر الضوئي المركب من الزخرفة السطحية شئ واضح وتظهر احياناً تجاعيد غامضة وبسيطة . وقد لاحظنا بمراقبة تدحرج حبات الطلع على الشريحة الزجاجية تحت المجهر انها ذات شكل شبيه بعضمه قصيرة ثلاثية الرأس ويمكن وصفها بـ Bone-like Pollen يكون احد الاقطاب احادي الفص حيث يتمثل القطب بقمه الفص والقطب الاخر يتمثل بالمنطقة المستوية او الضحلة المحصورة بين الفصين الاخرين .

للمدقة المفردة في الزهرة ميسم مفرد قرصي الى شبه كروي او شبه هرمي ، ابعاده بين 0.25-0.3 ملم وقلم مفرد انبوبي الشكل ابيض اللون خالي من الشعيرات ، طوله يتراوح بين 0.5-0.7 ملم وهو متاعي قاعي Gynobasic ومبيض ابيض اللون رباعي الفصوص شبه الكمثرية الشكل الملساء والذي تتراوح ابعاده بين 0.5-0.7 ملم .

يبدأ الكأس بالتضخم بعد التلقيح والاخصاب ويزداد التعرق الشبكي وضوحاً وبروزاً وتزداد مساحة سني الكأس الواقعين بين كل فصين متجاورين حيث تصل ابعاد الفص الواحد منها 4.5 x 4 ملم في جزئه المشطور فقط ، اما ابعاد الكأس الثمري ككل فهي 12 x 10 ملم ، ويكون الكأس الثمري المتوسع جلدي الطبيعة ويضغط على الثمرة بشدة حيث يزداد انضغاطه من الجهتين ، هذا ومما يميز هذا الجنس ونوعه بشكل جلي جداً هو شكل وطبيعة الكأس الزهري والكأس الثمري . وقد أشار Post (4) الى ان الكأس الثمري ينعقف متدلياً رغم قصر حامل الثمرة لكنه يبدو من ملاحظة النمائج العراقية في الدارسة الحالية ان هذه الصفة متميزة بهذا المعنى وربما ينعقف حامل الثمرة وليس دائما على جانب من جوانب المحور ، ورغم ذلك لاتتساقط بنيدقات الكأس الثمري بل تبقى داخل الكأس الثمري بعد اتمام نضجها ، وغالباً ما تصبح قمم الفصوص الثانوية في الكأس الثمري ثنائية القمم المحتدة Double acuminate apexes ، الشكل 3)-(2 وتتصلب شعيراتها اكثر ، هذا وتتغاير الشعيرات بتغاير البئة في اشكالها وكثافتها وتتغاير ايضاً على اعضاء الفرد النباتي الواحد . يضم الكأس الثمري الواحد اربع بنيدقات Nutlets عادة ، ابعاد الواحدة منها 3-3.5 x 2-2.3 ملم ذات لون بيج الى بني وشكلها بيضي Ovoid الى كمثري مضغوط الجانبين Pyriform compressed ، وتتصل كل بنيدقة Nutlet بانحامل الثمري Carpophore في ثلثها العلوي ومن جهتها البطنية ، وتظهر هذه البنيدقات وكأنها متصلة طولياً من جهاتها البطنية ، وهي ذات جدران صلبة جداً وسميكة ولها سطوح دقيقة الدرينات Minutely tuberculate الشكل (2-6- 7, 8) وذات قمة مقورة Obtuse وقاعدة مدورة عريضة ، اما السرة فتقع في قاعدة الثلث العلوي من الجهة البطنية وهي واضحة جداً مدورة الى اهليليجية الشكل .

ان طبيعة الكأس الثمري هذا واحتفاظه بالبنيدقات وطبيعة تلك البنيدقات هي تكيف ناجح لهذا النوع وتؤكد كونه نباتات ناجحة النمو والتكاثر في مختلف بيئات نموها . ورغم تميز نباتات هذا الجنس مظهرياً وانتشاره في الحقول والبئات التي يستعملها الانسان كالبساتين والمواقع التي يترك فيها مواشيه للرعي الا اننا لم نعثر له على اسماء محلية سوى ان بعض الفلاحين في بغداد يطلقون عليه كلمة لزيج نظراً لالتصاق نباتاته احيانا بالملابس وصوف الاغنام ولكنه يختلف كلياً عن نباتات الجنس Galium التي تسمى لزيج ايضاً نظراً لطبيعة التصاقها كذلك .

لقد وضع زهاري Zohary (17) هذا النوع A. Procumbens ضمن النباتات الشافية او التي تستعمل لشفاء المرضى والتآم الجروح والقروح . كذلك عده ضمن النباتات المستعملة كصبغات .

٢- البيئة والتوزيع الجغرافي Ecology and Geographical Distribution

من المعروف ان البيئة هي قاعدة التصنيف النباتي في فهم توزيع المراتب وتركيب الموسوعات

النباتية وفهم العلاقات التطويرية للمراتب التصنيفية والتغايرات في المجاميع السكانية . ان على باحث التصنيف ان يلاحظ الصفات المظهرية التي ترتبط مع العوامل البيئية المختلفة في بداية الامر وذلك من اجل فهم جيد للتغايرات التركيبية

في الحقل والمختبر ، كذلك عليه الربط بين مدى التغايرات البيئية وتاثيراتها على وصفه لمرتبة تصنيفية Taxon معينة ، رادفورد واخرون Radford et al.[18] . كذلك فان الالمام بالتوزيع الجغرافي واساسياته لاي جنس نباتي يساهم كثيرا في تحديد وعزل مراتب تصنيفية ادنى ضمن الجنس ، او حتى النوع ، ويعتبر ايضا كقاعدة في فهم الادلة النشوئية والتطورية واصل وهجرة الانواع او الاجناس .

ومن الملاحظات الحقلية ودراسة جميع ما تراكم من عينات للجنس في المعاشب العراقية ولعشرات من السنين فقد رسم صورة واضحة لطبيعة البيئة التي تنمو بها نباتات هذا الجنس في العراق.

تنمو نباتات الجنس Asperugo بشكل افراد او مجاميع سكانية قليلة العدد عادة على حافات الحقول او البساتين او الانهار او على جوانب احواض نباتات الزينة المستزرعة والغنية السقي ذات الترب الهشة عالية الرطوبة او الطينية بصورة عامة ، حيث نجد ان نباتات الجنس تفضل الترب الغرينية الرطبة ، كذلك تفضل الترب المضللة بالاشجار او بحافات الصخور ، وقد تنمو في الترب الرملية كما هي الحال في منطقة الصحراء الغربية والجنوبية او تنمو في الترب ذات الصخور المرمرية المحطمة كما في الجزيرة العليا وفي اثار الحضر . وقد تنمو في الترب الرطبة بين الصخور الجيرية كما هو الحال في مقاطعة جبل سنجار ، او قرب مساقط المياه في مقاطعة العمادية كما في العينة (34696BUH) ، اما من حيث الارتفاع فان نباتات الجنس ممكن ان تتواجد في مختلف الارتفاعات حيث توجد على مدى يتراوح بين ٢٠٠م فوق سطح البحر كما في مقاطعة الصحراء الغربية الى ٢٥٠٠م فوق سطح البحر كما هي الحال فوق جبل كاروخ.

ومن دراسة الخصائص المظهرية للجنس بصورة عامة نجد ان هناك ترابط وثيق بين الصفات التركيبية للنباتات ونوعية التربة او البيئة التي يعيش فيها ، حيث نجد ان السيقان الغضة والزاحفة في كثير من الاحيان تحتاج الى ترب ناعمة وهشه وذات رطوبة عاليه ، وهذا مالاحظناه في عموم عينات الجنس النامية في العراق .

اما من حيث التوزيع الجغرافي ، الشكل (٣) فتظهر نباتات الجنس Asperuge في معظم المقاطعات العراقية ، غير انها تكون اكثر كثافة في المقاطعات الجبلية وخاصة الغربية منها ثم تبدأ كثافتها تقل كلما اتجهنا الى شرق وجنوب العراق . ففي مقاطعة العمادية MAM ينتشر في كل من العمادية والمرتفعات المحيطة بها وفي كاني ماسي قرب الحدود التركية وفي عقره وناحية شريفة قرب العمادية وفي كلي زنطة بين عقره ودينارتا . اما في مقاطعة راوندوز MRO فتنتشر نباتاته في منطقة حاج عمران وقمم جبال راوندوز قرب خليفان وعلى جبل كاروخ وفي قرية وارته شمال غرب رانيه ، وعلى جبال زاينتا بين رانيه وشقلاوه ، اما في مقاطعة السليمانية MSU فتتواجد نباتاته على جبل قره داغ ثم تتجه الى اقصى الشمال الشرقي في بنجوين وعلى الطريق بين طويله وحلبجه وعلى سفوح جبل ازمر ثم يمتد انتشاره غربا الى مقاطعه جبل سنجار MJS حيث وجد على معظم السفوح الصخرية من الجبل وفي الوديان ذات الحافات والتربة الرطبه وخاصة قرب منابع او مجاري المياه ، اما في مقاطعة نينوى FNI فقد وجدت نباتاته في منطقة نينوى قرب الاثار وفي الحقول ، وقد جمع لاول مرة في هذه الدراسة من جبل مقلوب ضمن نفس المقاطعة ، ثم يتجه شرقا الى مقاطعة اربيل FAR حيث جمع من اسكي كلك وباستورا . ثم ينحدر في انتشاره جنوبا وباعداد قليلة جدا . ففي مقاطعة كركوك FKI ينتشر في الطوز ، وفي الجنوب الى اقصى الشرق في مقاطعة المرتفعات الحدودية الشرقية FFF وجد في منطقة السعدية. وتزداد كثافته قليلا في مقاطعة الجزيرة السفلى DLJ

حيث تنمو نباتاته في وقرب اثار الحضر وفي الجزء الغربي من هذه المقاطعة كذلك تتواجد قرب منطقة عنه ، واذا اتحدرنا جنوبا الى مقاطعة الصحراء الغربية DWD نجد ان نباتاته تنمو في منطقة كربلاء قرب حصن الاخيضر . اما في مقاطعة السهل الرسوبي الاوسط LCA فان انتشار نباتات الجنس يتركز في منطقة بغداد وخاصة في مناطق الكرادة والجادرية والكاظمية حيث المزارع والبساتين غزيرة المياه ، اما في مقاطعة الصحراء الجنوبية DSD فقد جمعت نباتاته من منطقة بين النجف وشبجة ومن البصية ، وبذلك ينقطع ظهور نباتات الجنس في اقصى الجنوب الشرقي من القطر ، الشكل (٣) ولم يتم جمع اية عينة من هذا النوع من باقي المقاطعات الجغرافية ، كما هو واضح من الخارطة في الشكل (٣) ، ويتوقع الباحثان ظهور هذه النباتات في هذه المقاطعات الاخيرة وخصوصا في المناطق المستزرعة منها بالنخيل والخضر .

وقد علمنا اخيرا ان نباتات هذا الجنس توجد بكثرة في بساتين النخيل في ابي الخصيب . وهنا تترك البيئة اثارها على المظاهر الخارجية للافراد النباتية وربما حتى على صفاتها المظهرية الدقيقة .

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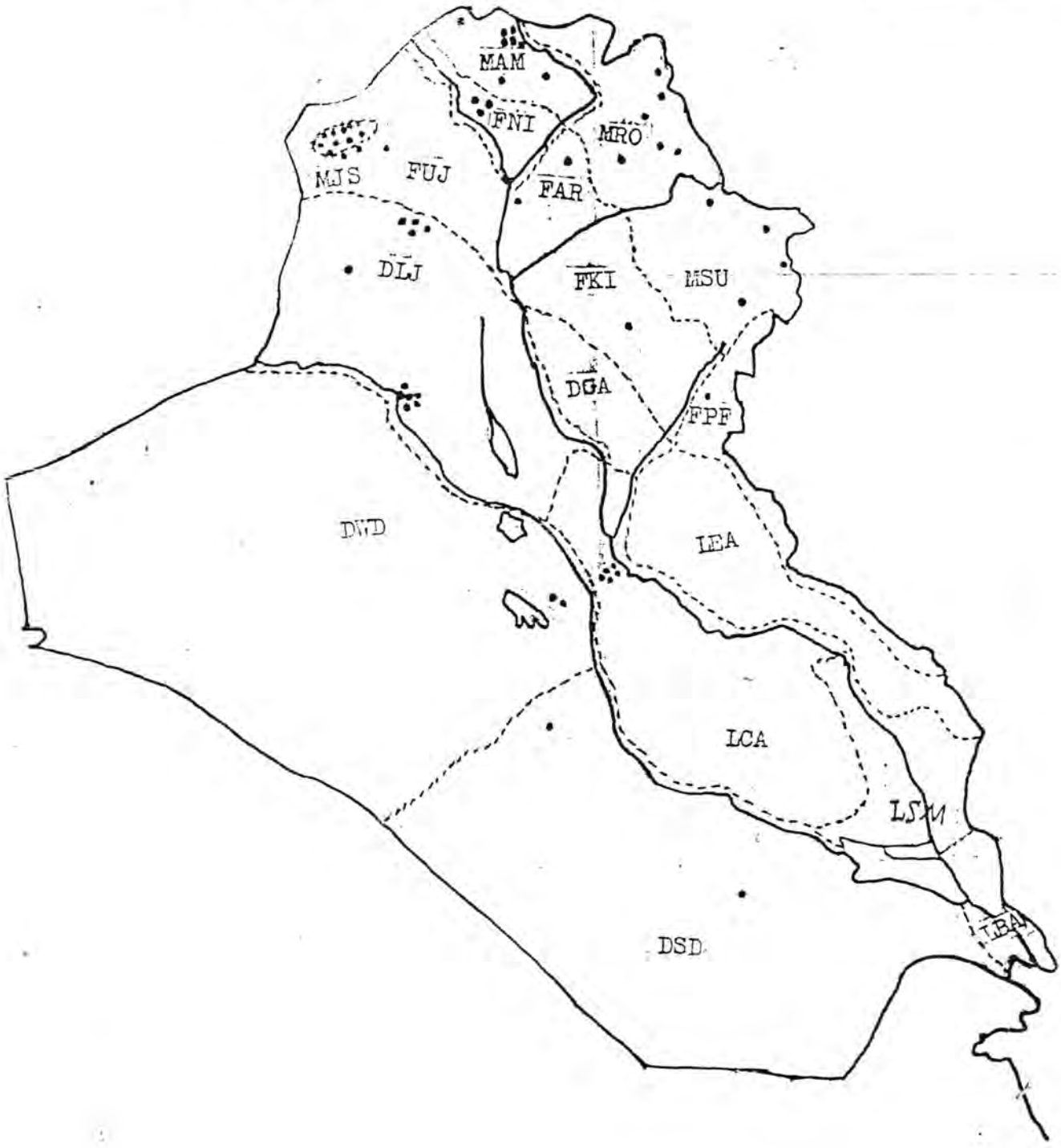
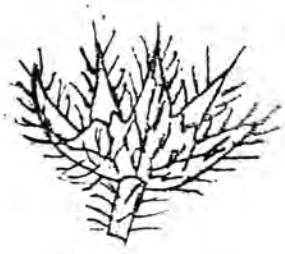
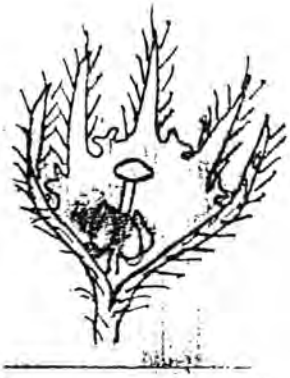


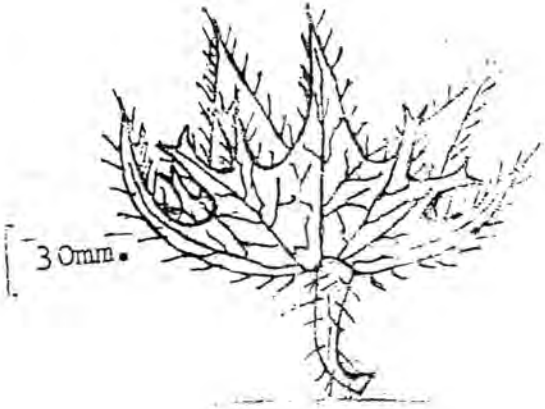
الشكل (٣) خارطة تبين توزيع الجنس Asperuge في المقاطعات الجغرافية العراقية



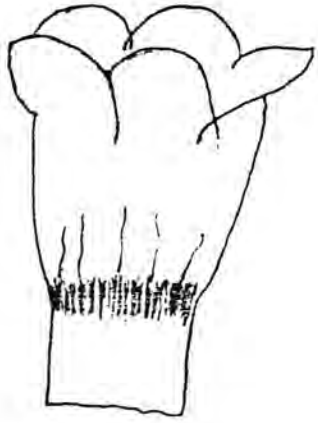
١- كاس زهري



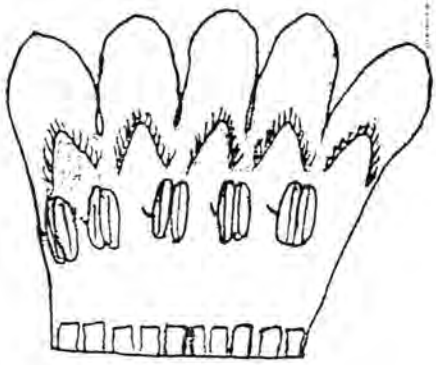
٢- كاس زهري مفتوح



٣- كاس ثمري

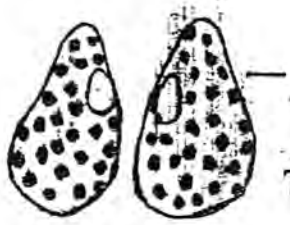


٤- تويج

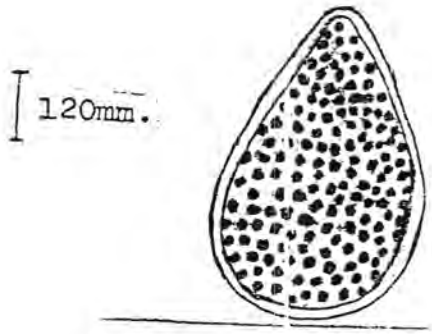


٥- تويج مفتوح

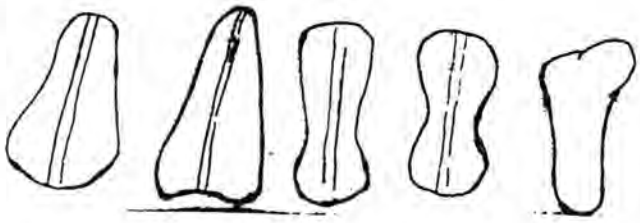
٦- اربع بنيدقات



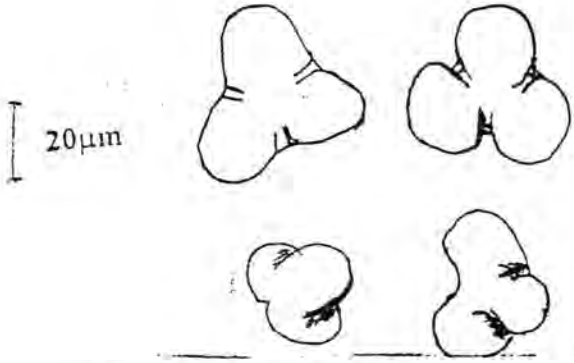
٧- الجهة البطنية للبنيدقة



٨- الجهة الظهرية للبنيدقة

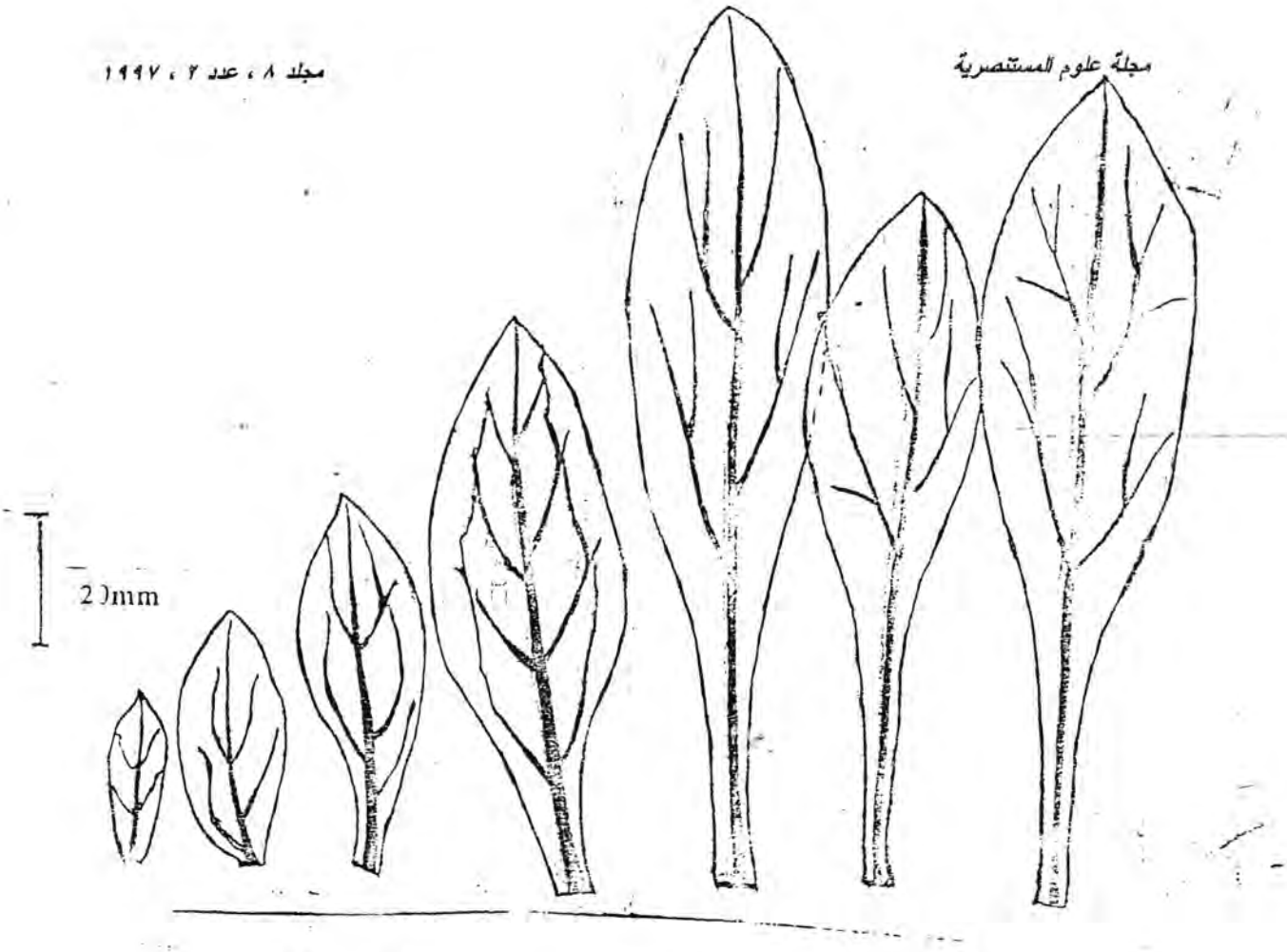


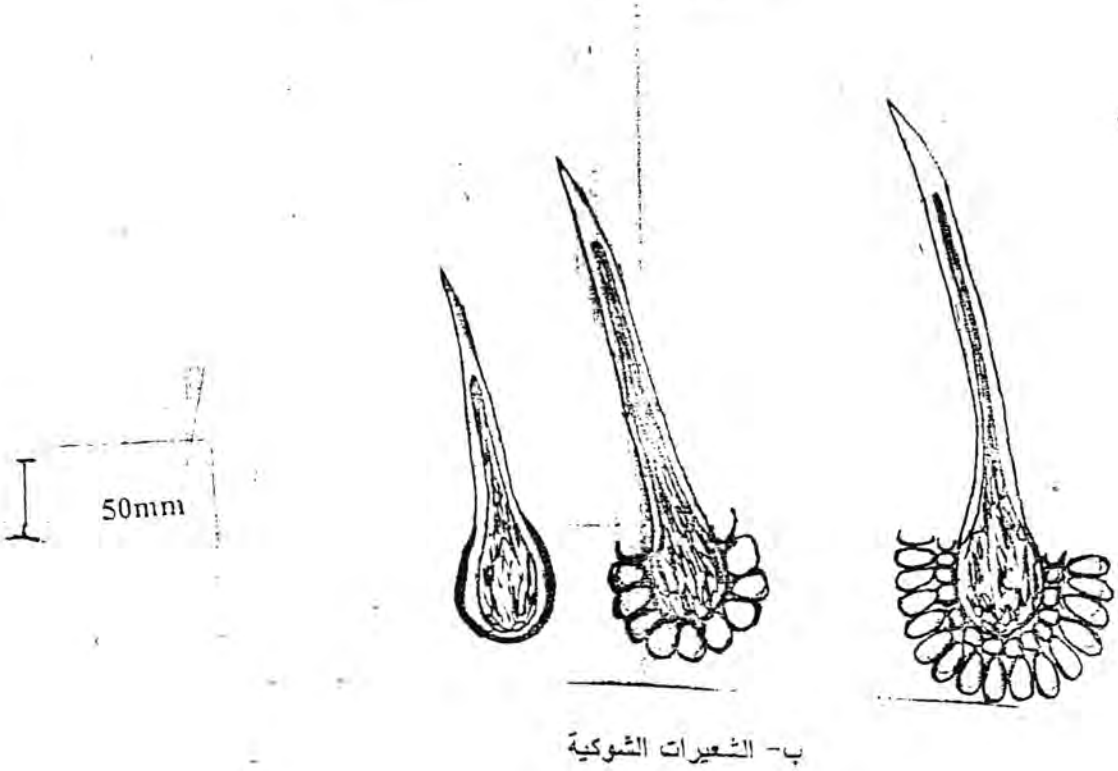
٩- منظر استوائي لحبوب اللقاح



١٠ منظر قطبي لحبوب اللقاح

الشكل (٢) بعض الاجزاء الزهرية والثمرية وحبوب اللقاح





شكل (١) الاوراق والشعيرات الشوكية

عزل وتشخيص الخمائر المتلبدة المنتجة للايثانول من البيئة المحلية

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ABSTRACT

Yeast isolates were recovered from local fruits after enrichement in Yeast extract medium with 10% ethanol. Fifty seven isolates were obtained with characteristics similar to yeasts of genus *Saccharomvces*. These yeast isolates were divided into five groups according to the degree of flocculation. Seven isolates were subjected to further characterization, six isolates (E_6, E_{15}, E_{37}, E_{42}, E_{58}, E_{64}) were found similar to *S. cerevisiae*, while one isolate (E_{70}) was similar to *S. uvarum*. The level of flocculation was studied in correlation with ethanol production. Isolates E_6 and E_{42} with weak flocculation property produced 4.7% and 4.5% (v/v) ethanol respectively. Whereas, yeast isolate E_{64} with moderately flocculation production 4% (v/v) ethanol. On the other hand, isolate E_{70} with good flocculation produced 4.3% (v/v) ethanol as compared with ethamol production of standard strain *S. uvarum* produced 3.9% (v/v) ethanol. These results suggest the possibility of isolating goos floccculent yeast with good ability to produce ethanol from local habitat.

الخلاصة

عزلت سلالات خميرة من الفاكهة المحلية المتضررة ، بعد تنميتها في وسط Yel الذي يحتوي على ١٠٪ ايثانول وتم الحصول على سبعة وخمسين عزلة تمتلك نفس المواصفات المظهرية والمجهرية لجنس Saccharomyces وقسمت هذه العزلات الى خمس مجاميع اعتمادا على درجة تلبدها وبينت الدراسة التشخيصية لسبعة عزلات منتقاة متباينة في درجة تلبدها ، ان ست عزلات (E_{64}, E_{58}, E_{42}, E_{37}, E_{15}, E_6) كانت مماثلة لمواصفات لخميرة الخبز القياسية S. cerevisiae وعزلة واحدة فقط (E_{70}) مطابقة للخميرة القاعدية المتلبدة S. uvarum اختيرت اربع عزلات بشكل عشوائي وبمستويات تلبد مختلفة ثلاثة منها تمثل S. cerevisiae واخرى تمثل S. uvarum لدراسة انتاجيتها للايثانول ، ولوحظ ان العزلة الضعيفة التلبد E_6 انتجت ٤,٧٪ (ح/ح)ايثانول ، بينما بلغت نسبة الايثانول المنتج من قبل العزلة E_{42} القليلة التلبد ٤,٥٪ (ح/ح) ،في حين اظهرت العزلة E_{64} المتوسطة التلبد انتاجية منخفضة للايثانول بحدود ٤٪ (ح/ح). وقد تميزت العزلة E_{70} ذات التلبد الجيد انتاجية جيدة للايثانول بلغت ٤,٣ (ح/ح) مقارنة مع السلالة القياسية المتلبدة S. uvarum التي اعطت ٣,٩٪ (ح/ح) ايثانول . يتضح من نتائج هذا البحث امكانية عزل خمائر جيدة التلبد من البيئة المحلية تمتلك قدرة جيدة على انتاج الايثانول .

المقدمة

اتجه المعنيون بالصناعات التخمرية للبحث عن سلالات خمائر تمتلك خاصية التلبد Flocculation، لدورها الفاعل في حل احدى المشاكل التي تواجه هذه الصناعة الا وهي جمع الخلايا وفصلها عن المنتوج النهائي بعد انتهاء عملية التخمر دون الحاجة الى استخدام اجهزة خاصة لهذا الغرض[٢،١]. تتميز خلايا الخمائر المتلبدة بقابليتها على التجمع الذاتي وبشكل كتل سرعان ما تنفصل عن الوسط السائل الذي تعلق فيه عند بلوغها نهاية الطور اللوغارتمي من النمو لتستقر في اسفل الوعاء التخمري[٣]، وتعود اليه التلبد في الخمائر الى التجاذب الايوني والتداخلات الكيميائية للمجاميع والشحنات الموجودة على سطوح خلاياها[٤].

اشارت بعض الدراسات الى انتشار ظاهرة التلبد بشكل واسع وبدرجات مختلفة في خميرة البيرة من نوع *Saccharomyces uvarum* و *S. carlsbergensis* كما ولوحظ تواجد هذه الظاهرة في بعض سلالات خميرة الخبز *S. cerevisiae* [٦،٥]، علما بان كلا الخميرتين تعد من الخمائر المنتجة للايثانول ، وسبق ان اشارت الدراسات المنشورة الى ان الخمائر المتلبدة اقل كفاءة في انتاج الايثانول [٧]، لذا فان البحث عن سلالة تمتلك خاصية التلبد وذات انتاجية عالية للكحول سيؤدي الى تطوير تطبيقات التقانات الحياتية في انتاج هذه المادة . وحديثا اجريت دراسات حول تواجد الخمائر المتلبدة في البيئة العراقية [٨] ولم تشر الدراسات الى عزل الخمائر المتلبدة المنتجة للايثانول من البيئة المحلية ، وعلى ضوء ذلك فقد هدف البحث الى التعرف عن امكانية عزل سلالات خميرة متلبدة ذات انتاجية عالية للكحول الاثيلي من البيئة المحلية .

المواد وطرائق العمل

الخمائر القياسية : استخدمت سلالتان من خمائر المقارنة القياسية وهما : خميرة الخبز غير المتلبدة من نوع *S. cerevisiae* والخميرة القاعية المتلبدة من نوع *S. uvarum* .

العزلات المحلية : اتبعت طريقة Ibrahim et al. (٩) لعزل وانتقاء الخمائر بتنميتها على الوسط Yeast Extract (١٠) والذي يحتوي على ١٠٪ ايثانول وتمت تنقية العزلات على وسط Yeast Extract (YEA) وحفظت عزلات الخمائر اما على وسط مائل YEA أو وسط YE بعد اضافة الكليسرول (١٠) .

توصيف العزلات : اتبعت الطرائق التي ورد ذكرها في Lodder (١١) لتشخيص العزلات ونوعها ، اذ شملت دراسة للصفات المظهرية والمجهرية والصفات الفسيولوجية ، التي تضمنت اختبار قابليتها في تخمير وتمثيل السكريات وتمثيل المصادر النايتروجينية وحساسيتها للسايكلوهكساميد .

قياس ظاهرة التلبد : قيست ظاهرة التلبد لعزلات الخمائر المحلية (مع سلالتي المقارنة) النامية في وسط منقوع الشعير Wort 12° (١٢) باستخدام طريقة حجم الراسب التي اوردها Thorne (١٣) وفيها جمعت خلايا الخميرة لكل عزلة بواسطة جهاز الطرد المركزي وغسلت بالماء المقطر وعلق ١غم من خلايا كل عزلة على انفراد في ١٠ مللتر من منظم الخلات برقم هيدروجيني ٤،٥ والمحتوي ٠،٠٥٪ كبريتات الكالسيوم ومزجت الخلايا جيداً ، ثم حسب حجم الراسب بعد مرور ١٠ دقائق باستعمال انابيب مدرجة من صفر الى ١٠ مل ووصفت درجة التلبد للخمائر استناداً الى حجم الراسب .

وسط التخمير وانتاج الايثانول : لقح وسط التخمير (٧) بـ ١٠٪ من اللقاح النامي في وسط YE بعمر يومين ودرجة حرارة ٣٠ م$^{\circ}$. ثم عرض ويط التخمر للتهوية لمدة ست ساعات في درجة حرارة ٣٠ م$^{\circ}$ ، بعد ذلك

اوقفت التهوية لبدء عملية التخمر لمدة ٤٨ ساعة . واخذت نمائج وسط التخمر لغرض تقدير السكر استناداً الى طريقة Milter (١٤) وتقدير الكحول الاثيلي المنتج بالطريقة الحجمية باستعمال Pyconometer متبعا للمرجع (Method of analysis of the emerican society of brewing chemist, 1982)

التحليل الاحصائي : تم تحليل النتائج احصائياً اعتمادا على اختبار t (t-test) (١٥) .

النتائج والمناقشة

تم الحصول على ثلاث وسبعين عزلة خميرة من الفاكهة المتخمرة بعد زرع نماذج منها على وسط YEL الحاوي على ١٠٪ ايثانول ، وتبين ان سبع وخمسين عزلة منها تمتلك مواصفات مظهرية ومجهرية مطابقة لجنس Saccheromyces . قسمت هذه العزلات الى مجاميع اعتماداً على درجة تلبدها (جدول ١) فالمجموعة الاولى (عديمة التلبد) ضمت ٨٧،١٪ من العزلات وشملت المجموعة الثانية (ضعيفة التلبد) ٥،٢٪ من العزلات واحتوت الثالثة (قليلة التلبد) على ٣،٥٪ من العزلات ، في حين بلغت النسبة المئوية لتواجد عزلات كل من المجموعة الرابعة (متوسطة التلبد) والمجموعة الخاصة (جيدة التلبد) ١،٧٪ من المجموع الكلي للعزلات . كما ويوضح الجدول (٢) نتائج فحص الصفات الفسيولوجية لسبع عزلات متباينة في درجة تلبدها (مقارنة مع السلالتين القياسية) فقد كانت نتائج العزلات الست (E_6 ، E_{15} ، E_{37} ، E_{42} ، E_{58} ، E_{64}) مطابقة لخميرة الخبز القياسية *S. cerevisiae* اذ اعطت فحصاً موجباً في تخمير وتمثيل كل من السكريات : الكلوكوز والكالاكتوز والمالتوز والسكروز والرافينوز وفحصاً سالباً في تخمير وتمثيل كل من سكر اللاكتوز والليليبوز والسليبايوز والنشأ ، بينما اظهرت عزلة واحدة فقط (E_{70}) نتائج مماثلة للخميرة القاعية المتلبدة *S. uvarum* ، اذ تمكنت من تخمير وتمثيل كل من سكر الكلوكوز والكالاكتوز والمالتوز والسكروز والرافينوز اضافة الى المليبايوز وفشلت في تخمير وتمثيل بقية السكريات . وسبق أن أكد Hough (١٦) على اهمية اجراء اختبار قابلية الخميرة على تخمير وتمثيل سكر المليبايوز كأساس للتفريق بين العزلات التابعة للـ *S. cerevisiae* و *S. uvarum* ، كما ويبين الجدول نفسه عدم امكانية العزلات السبع (مع سلالتي المقارنة) على استغلال النترات والنتريت كمصدر للنتروجين ، اضافة الى حساسيتها للسايكلوهكساميد ، اذ عدت الحساسية المفرطة لهذا المضاد الحيوي وبتركيز (١ مايكروغرام / مل) أحد الصفات المعتمدة لتشخيص خميرة الخبز والخميرة القاعية (١١،١٧) .

تم انتقاء اربع عزلات خميرة بمستويات مختلفة من التلبد وبشكل عشوائي بهدف تقييم انتاجيتها للايثانول ، ومن خلال النتائج الموضحة في الجدول (٣) تبين ان انتاج العزلة E_6 *S. cerevisiae* الضعيفة التلبد للايثانول ٤،٧٪ (Z/Z) قد قاربت انتاجية خميرة الخبز القياسية للايثانول التي اعطت ٤،٩٪ (Z/Z) ، في حين بلغت انتاجية العزلة E_{42} *S. cerevisiae* (قليلة التلبد) للايثانول ٤،٥٪ (Z/Z) وأكدت نتائج التحليل الاحصائي الى عدم وجود فروق معنوية في انتاجية هاتين العزلتين للايثانول مقارنة مع السلالة القياسية *S. cerevisiae* بالرغم من وجود اختلاف ملحوظ في نتائجهما . بينما اظهرت العزلة المتوسطة التلبد E_{64} *S. cerevisiae* انتاجية منخفضة للايثانول بلغت ٤٪ (Z/Z) وبفارق معنوي يساوي ٩ عند قياسه بمستوى ٠،٠٥ وقد تميزت العزلة الجيدة التلبد E_{70} *S. uvarum* بانتاجية جيدة للايثانول بمقدار ٤،٣ (Z/Z) مقارنة مع انتاجية الخميرة القاعية المتلبدة *S. uvarum* للايثانول حيث كانت بحدود ٣،٩٪ (Z/Z) ولكن لم يشر التحليل الاحصائي الى وجود فروق معنوية في انتاجية هذه العزلة للايثانول ومن الجدير بالذكر ملاحظة كمية السكر المتبقي المثبتة في الجدول نفسه الذي يعد كمؤشر على

كفاءة تخمير السكر الى الايثانول ، ففي حالة وجود كمية من السكر المتبقي سيكون دليلاً على عدم كفاءة التخمير مقارنة مع العزلات التي تستهلك السكر بكفاءة عالية ، وفي هذا المجال اشار Phaff et al. (١٨) الى ان هذه العمليات الحيوية تعتمد على الانظمة الانزيمية لتحليل السكر ونقله الى داخل الخلية بواسطة انظمة نقل معينة ومن ثم تقوم خلايا الخميرة بتحويل ٧٠٪ منه الى غاز ثاني اوكسيد الكاربون وايثانول .

يتضح مما تقدم ، وجود علاقة بين درجة تلبد الخمائر وانتاجيتها للايثانول ، اذ ان الخميرة القاعية المتلبدة (القاياسية) ذات انتاجية منخفضة مقارنة مع انتاجية سلالات عديدة من خميرة الخبز ولكن اوضحت هذه الدراسة امكانية عزل الخميرة ذات نمط مظهري جيد بخاصية التلبد وتمتلك في نفس الوقت انتاجية اعلى من الخميرة القاعية القياسية ، وعموماً اكدت النتائج المذكورة في هذا البحث الى اهمية الاستمرار والتوسع في التحري عن علاقة التلبد والانتاجية للايثانول وذلك بانتخاب سلالات ذات خصائص معينة بدرجات التلبد لاهمية التلبد في مجمل الصناعات التخميرية . كما وتجدر الاشارة هنا الى امكانية استخدام طفرات وراثية امامية وراجعة لانتقاء سلالات الاكثر ملائمة من حيث انتاجيتها للكحول الاثيلي وسرعة ترسبها (١٩) . وعليه يمكن الاستنتاج من خلال هذه الدراسة جدوى البحث عن الخمائر المتلبدة والمنتجة للايثانول من البيئة المحلية مع اهمية استخدام الطرائق الوراثية لتطويرها في هذا المجال .

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جدول (١) : مجاميع عزلات الخميرة المحلية مصنفة حسب درجة تلبدها

رقم المجموعة	درجة التلبد للعزلات المحلية	عدد العزلات	حجم الراسب (مل)	النسبة المئوية لتواجد العزلات
١	عديمة التلبد	٥٠	اقل من ٠،١	٨٧،١
٢	ضعيفة التلبد	٣	٠،٥	٥،٢
٣	قليلة التلبد	٢	٠،٨	٣،٥
٤	متوسطة التلبد	١	١،٠	١،٧
٥	جيدة التلبد	١	١،٨	١،٧

الخمائر القياسية	درجة التلبد	حجم الراسب
S. cerevisiae	عديمة التلبد	صفر
S. uvarum	جيدة التلبد	٢

ملاحظة :

عديمة التلبد =صفر-٠،١ مل من حجم الراسب

ضعيفة التلبد =٠،٢-٠،٦ مل من حجم الراسب

قليلة التلبد =٠،٧-٠،٩ مل من حجم الراسب

متوسطة التلبد=١،٠-١،٥ مل من حجم الراسب

جيدة التلبد =١،٦ فما فوق مل من حجم الراسب

جدول (٢) : الفحوص الفسيولوجية لعزلات الخميرة المنتقاة بمستويات من التلبد

سلالات الخمائر	القابلية على تخمير المصادر الكاربونية وتمثيلها									تمثيل المصادر النايتروجينية		المقاومة للسايكلوهكساميد
	كلوكوز	كالاكتوز	مالتوز	سكروز	لاكتوز	رافينوز	ملبيايوز	سليبايوز	نشأ	النترات	النتريت	
أ- الخمـائر المماثلـة لخميرة الخبز												
- الخميرة القياسية : *S. cerevisiae*	+	+	+	+	−	+	−	−	−	−	−	−
-العزلات المحلية : (E_6، E_{15}، E_{37}، E_{42})	+	+	+	+	−	+	−	−	−	−	−	−
(E_{58}، E_{64})												
ب- الخمـائر المماثلـة للخميرة القاعية												
- الخميرة القياسية *S. uvarum*	+	+	+	+	−	+	+	−	−	−	−	−
-العزلة المحلية : E_{70}	+	+	+	+	−	+	+	−	−	−	−	−

+ ظهور النمو

− عدم ظهور النمو

جدول (٣) : انتاج الايثانول وتقدير السكر لعزلات الخميرة المحلية ذات درجات تلبد متباينة

العزلات المحلية	درجة التلبد	النسبة المئوية للسكر المتبقي	النسبة المئوية للايثانول المنتج
S. cerevisiae E_6	ضعيفة التلبد	٠,٥	٤،٧
S. cerevisiae E_{42}	قليلة التلبد	٠،٧	٤،٥
S. cerevisiae E_{64}	متوسطة التلبد	١،٧	٤،٠
S. uvarum E_{70}	جيدة التلبد	١،٥	٤،٣
الخمائر القياسية			
S. cerevisiae	عديمة التلبد	صفر	٤،٩
S. uvarum	جيدة التلبد	٢	٣،٩

تأثير الزئبق اللاعضوي على المقاومة الطبيعية والوراثية للبكتريا الموجبة والسالبة لصبغة غرام المحلية

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ABSTRACT

To ascertaiin the antibacterial activity of the non-organomercuial compound ($HgCl_2$), the M.I.C (minimum inhibitaty concentration) was used in liquid and solid media. Two isolated of each *E. coli* and *Staph. aureus* were used as indicaters for Gram begative and Gram posative bacteria. Results obtained indicated that *E. coli* was naturelly more resistant than *Staph. aureus*. Resistant mutants isolated on nutrient agar showed that the two isolates of *E. coli* were resistant, while only one isolate of *S. aureus* showed resistance.

الخلاصة

درست مقاومة البكتريا الموجبة والسالبة لصبغة غرام للمادة الفلزية اللاعضوية (كلوريد الزئبق $HgCl_2$) من خلال قياس التركيز الادنى المثبط لنمو عزلتين من العنقوديات الذهبية وعزلتين من بكتريا الايشيريشيا القولونية باستخدام وسط المرق المغذي (وسط سائل) ووسط الاجار المغذي (وسط صلب كل على حدة . لقد اظهرت نتائج البحث بان بكتريا العنقوديات الذهبية *S. aureus* تمتلك حساسية ملحوظة تجاه المادة السامة اذا تراوح التركيز الادنى المثبط للنمو بين ١,٥-٢ مايكروغرام /سم^3 في الوسط السائل و ٢,٥-٣,٥ مايكروغرام /سم^3 في الوسط الصلب - وبالمقارنة فان بكتريا الايشيريشيا القولونية *E. coli* ابدت مقاومة طبيعية عالية نسبيا وباستخدام الوسطين السابقين . وبينت الدراسة امكانية عزل طفرات وراثية مقاومة على وسط الاجار المغذي من عزلتي الايشيريشيا القولونية ومن عزلة واحدة من العنقوديات الذهبية .

المقدمة

تتصف مركبات الزئبق بقابليتها على قتل البكتريا والفطريات [١]. تعتمد هذه المركبات في ميكانيكيتها على ايوناتها التي تتحد مع مجاميع الثايول ، حيث يكون الناتج مركب معقدا يسمى الميركابتايد[٢] وبذلك تمنع التخليق الحيوي للبروتينات وعملية استنساخ المادة الوراثية . وعرفت مقاومة البكتريا الموجبة والسالبة لصبغة غرام لمركبات الزئبق [٤،٣] وعزيت المقاومة الى البلازميدات وعلى وجه الخصوص في البكتريا السالبة لصبغة غرام[٥]. وقد تاتي المقاومة نتيجة طفرات كروموسومية[٧،٦]. وتحدث المقومة كنتيجة لاختزال Hg^{+2} الى Hg^{o} الاقل سمية[٨].

المواد وطرائق العمل

١- محلول كلوريد الزئبق $HgCl_2$ (Fluka)

٢- عزلتان محليتان من بكتريا العنقوديات الذهبية Staphylococus A12&B12 تم الحصول عليهما من خزين قسم علوم الحياة / كلية العلوم / الجامعة المستنصرية .

واستعملت عزلتان لبكتريا الايشيريشيا القولونية العزلة الاولى *E. coli* K69 تم الحصول عليها من المصدر (٩).

اما العزلة المحلية الاخرى *E. coli* N17 فقد تم الحصول عليها من قسم علوم الحياة / كلية العلوم/ الجامعة المستنصرية .

٣- قياس التركيز الادنى المثبط للنمو .

أ- في وسط المرق المغذي : استخدمت طريقة Bliss et al (١٠) وتتم باضافة تراكيز متدرجة من محلول كلوريد الزئبق الى انابيب الاختبار المحتوية على ٥سم^{٣} من المرق المغذي (مضاعف التركيز) ويكمل الحجم الى ٩,٩ سم^{٣} بالماء المقطر المعقم. تلقح جميع انابيب التجربة بـ او . سم^{٣} من مزروع البكتريا وبعمر ١٦-١٨ ساعة. تتابع النتائج لمدة خمسة ايام من الحضانة في ٣٧° م وتقارن بانابيب السيطرة .

ب- في وسط الاجار المغذي :

تستعمل لهذا الغرض اطباق بترى محتوية على الاجار المغذي المزود بتراكيز متدرجة من محلول $HgCl_2$. يتم زرعها بحجم ٠,٢ سم^{٣} من مزروع البكتريا وبعمر ١٦-١٨ ساعة وتتابع النتائج لمدة سبعة ايام من الحضانة في ٣٧° م وتقارن باطباق السيطرة .

٤-الحصول على الطفرات الوراثية المقاومة

استعملت البكتريا في طور الثبوت وزرعت بمعدل ١٠x٣ ^{٩} خلية في كل طبق يحتوي على اقل من كلوريد الزئبق لايعطي نموا مقارنة لنمو السيطرة ، واعتمدت النتائج بعد سبعة ايام في ٣٧° م .

النتائج والمناقشة

اشارت النتائج المذكورة في الجدول (١) الى حدود المقاومة الطبيعية للعزلات الاربعة حيث تبين ان العزلة *E. Coli* K69 تمتلك اعلى حد للمقاومة وهي بحدود ٥ مايكروغرام / سم^{٣}، في حين كانت المقاومة الطبيعية للعزلة *S. aureus* B12 بحدود ١,٥ مايكروغرام / سم^{٣} وهذه النتائج تتفق مع ما جاء به (١١)، اذ ان المقاومة الطبيعية للبكتريا السالبة لصبغة غرام هي اعلى من تلك التي تمتلكها البكتريا الموجبة لصبغة غرام.

واكدت النتائج المذكورة في الجدول (٢) بان العزلة *E. coli* K69 تمتلك اعلى حد لمقاومة الطبيعية في وسط الاجار المغذي وهي بحدود ٧ مايكروغرام / سم^{٣} .

ان هذه المقاومة العالية نسبيا للعزلة *E. coli* K69 قد يعزى الى كونها Entrotoxigenic (٩) . اما العزلة *S. aureus* B12 تمثل اقل حد للمقاومة الطبيعية وهي بحدود ٢,٥ مايكروغرام / سم^{٣} . ان لطبيعة الوسط الزراعي دور مهم يؤدي الى اختلاف في النتائج كما هو واضح في الجدول (١) والجدول (٢) .

وعلى ضوء نتائج التركيز الادنى المثبط لنمو العزلات البكتيرية المستعملة في الدراسة فقد تم التحقق من امكانية حدوث طفرات مقاومة لكلوريد الزئبق ومدى التكرار الطفوري . ولقد اشارت النتائج المدونة في الجدول (٣) الى ان التكرار الطيفوري للعزلة *E. coli* K69 بحدود ١٠x١٣ $^{-١٠}$ طفرة خلية مقارنة بـ ١٠x١٦ $^{-١٠}$ طفرة /خلية بالنسبة للعزلة *E. coli* N17 و *S. aureus* A12 ، اما العزلة *S. Aureus* B12 فان تكرارها الطفوري هو صفر لانها فشلت في مقاومتها للمادة السامة . ان مقاومة الاحياء المجهرية للزئبق ومركباته اشارت اليها الكثير من البحوث (٤)، وتاتي هذه المقاومة عن طريق البلازميدات والكروموسومات النووية (٦)، وربما تعزى مقاومة

العزلات الثلاث الاولى الموضحة في الجدول (٣) الى حصول طفرات كروموسومية او بلازميدية ، وبهدف التحقق من ذلك يتطلب اجراء دراسات بايولوجية جزيئية للكشف عن دور البلازميدات في منح المقاومة في هذه العزلات .

جدول ١ تاثير الزئبق اللاعضوي في وسط المرق المغذي على عزلات بكتريا *S. aureus & E. coli*

العزلة البكتيرية	العدد الحي للخلايا المزروعة ١٠X ^{٦}سم^{٣}	تركيز الزئبق اللاعضوي المثبط للنمو مايكروغرام / سم^{٣}
E. coli K69	٣,٧	٥
E. coli N17	٣,٨	٤
S. aureus A12	٣,٩	٢
S. aureus B12	٣,٧	١,٥

جدول ٢ تاثير الزبق اللاعضوي في وسط الاجار المغذي على عزلات الـ*E. coli* والـ*S. aureus*

العزلة البكتيرية	العدد الحي للخلايا المزروعة ١٠X ^{٩}سم^{٣}	تركيز الزبق اللاعضوي المثبط للنمو مايكروغرام/سم^{٣}
E. coli K69	١,٩	٧
E. coli N17	٢,٠	٥
s. aureus A12	٢,١	٣,٥
S. aureus B12	٢,١	٢,٥

جدول رقم ٣ التكرار الطفوري للمستعمرات المقاومة للزئبق اللاعضوي لعزلات البكتريا الـ *E.coli* و *S. aureus*

عزلة البكتريا	العدد الحي المزروع ١٠X ^{٩}سم^{٣}	تركيز الزئبق مايكروغرام/ سم^{٣}	عدد الطفرات	التكرار الطفوري طفرة / خلية
E. coli K69	٣	٧	٤	١٣ ١٠X $^{-١٠}$
E. coli N17	٣,٢	٥	٢	١٦ ١٠X $^{-١٠}$
S.aureus A12	٣,٣	٣,٥	٢	١٦ ١٠X $^{-١٠}$
S.aureus B12	٣,١	٢,٥	صفر	صفر

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