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PII: S0308-8146(98)00005-3

Food Chemistry, Vol. 63, No. 4, pp. 535–541, 1998

Printed in Great Britain
0308-8146/98 $19.00 + 0.00

Nutrients and chemical residues in an Egyptian total mixed diet

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(Received 25 July 1997; accepted 13 October 1997)

A mixed diet composite (EGYPT-DIET95) was prepared to represent the intake of Egyptian urban adults. Proximate analyses and assays for selected toxic elements, as well as organic chloro compounds, were carried out on this composite using well established methods and certified RMs (reference materials). The analysis of the composite diet demonstrates its usefulness for assessment of daily intakes. Calculated daily intakes of toxic substances were compared with the Acceptable Daily Intake of Codex Alimentarius (1984). The amounts of mercury and nitrates in Egyptian total-diet samples are of health concern. © 1998 Elsevier Science Ltd. All rights reserved.

INTRODUCTION

Total diet studies (TDS), as defined and recommended by the FAO/WHO (WHO, 1976, 1985), have been carried out for many years in the UK (Peattie *et al*., 1983), the USA (Iyengar *et al*., 1987) and the Netherlands (Van Dokkum *et al*., 1989). The aims of total diet studies are to evaluate the nutritional quality of 'national diets', and to monitor the exposure to additives and contaminants through habitual diets and to estimate the health risk for the consumer, by comparing contents with the acceptable daily intakes (ADI) established by the Food and Agriculture Organization/World Health Organization (FAO/WHO). A further objective of the TDS is to observe trends in the consumption of those analytes over time (Iyengar *et al*., 1987).

A research program on dietary intake of nutrients and other elements among adult Egyptians has been initiated by our Department (Hussein *et al*., 1995, 1997).

The objective of the present investigation is to report the procedural details of the preparation of the first Egyptian benchmark mixed diet, and to report analytical data regarding proximate analysis and fatty acid profiles as well as heavy metals, nitrates and chlorinated pesticide residues. The composition data, together with food consumption data, enable calculation of average daily intakes.

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MATERIALS AND METHODS

Collection and preparation of the mixed diet

The plan for the collection of food in this project followed the Joint FAO/WHO Guidelines (1985) for the study of dietary intakes of chemical contaminants. Briefly, this plan was based on the approach of the total diet study (TDS), in which a market basket of foods reflects a defined total diet of a consumer for a specific period of time; the foods are prepared for table-ready consumption.

The mixed diet was prepared from 76 individual foods representing those consumed by a population group 23–65 years of both sexes living in the governorates of Cairo and Giza.

The approach adopted in the food consumption survey was sampling for three days using 24 h recalls and/or food diaries. The diets collected were intended for subjects with no special dietary requirements and provide 2800 kcal (Hussein *et al*., 1995). The programme involves the development of food lists defining the quantities of commonly consumed foods whose daily consumption is 0.1 g or more (Table 1). Foods were collected from four different districts representing different socioeconomic classes of Cairo and Giza governorates. In each district, all the foods were collected according to the same shopping guide; enough foods were purchased to represent 3 total daily diets in terms of amount of food.

Table 1. Amounts of selected food commodities used for the preparation of the mixed diet

Food	Moisture %	Mean estimate for daily intake (g)
Cereals		
Bread, wheat flattened, 85% extr.	24.6	202.2
Bread, wheat flattened, 72% extr.	26.3	0.4
Bread, wheat rolls, 72% extr.	24.8	14.4
Bread, wheat buns, 72% extr.	21.4	2.0
Wheat grains, cooked (belila)	87.2	0.85
Wheat flour	10.6	1.56
Macaroni, cooked	38.8	25.1
Macaroni, lassagne	52.5	3.0
Pizza	33.5	1.2
Bars, rings, zwieback	11.5	5.3
Rice, cooked	58.8	198.0
Sweets		
Biscuits	4.0	4.9
Chocolates	2.9	0.8
Honey	13.3	2.8
Kekes, raisin, 3 brands	27.8	6.0
Oriental puff paste, heavy syrup	21.1	0.85
Puff paste, home made, sugar	6.1	3.2
Basbousa, heavy syrup	13.0	0.8
Waffles filled, 2 brands	8.0	1.64
Sugar cane	0.2	56.2
Molasses	21.3	1.2
Tubers		
Potatoes cooked	69.4	47.9
Potatoes chips	40.0	4.6
Taro, cooked + leaf celery	73.4	3.1
Vegetables		
Cabbage leaves, stuffed with rice	83.8	0.3
Carrots, fresh + cooked	89.4	33.3
Cauliflower, cooked, tomatoes	65.0	0.54
Cucumber	95.1	3.5
Eggplant, cooked	80.1	0.24
Garlic	62.1	1.1
Okra, cooked, tomatoes	72.0	2.5
Olives, black	72.0	0.6
Olives, green	72.6	0.2
Onions	90.7	16.2
Tomatoes	94.8	7.5
Tomatoes canned	69.2	6.7
Vegetable soup	88.5	98.0
Leafy vegetables		
Spinach, tomatoes	76.5	3.1
Lettuce, fresh	94.9	1.8
Parsley	83.3	5.0
Raddish, white, fresh	90.3	2.0
Garden rocket, Eruca sativa	89.7	19.0
Fruits		
Apple	81.3	1.5
Mandarin	83.0	5.1
Orange	84.8	10.9
Orange juice	89.4	0.34
Strawberry	89.8	0.34
Legumes		
Faba beans, stewed	72.3	28.0
Beans, white, tomatoes	69.5	5.7
Lentils	83.5	4.6
Peanuts	3.8	0.8
Peas, tomatoes	72.3	19.6
Sesame	4.4	3.7
Sesame tehena	0.7	1.4
Cowpeas, cooked, tomatoes	75.0	4.6
Water melon seeds	1.5	7.9
Meats and fishes		
Eggs, chicken, boiled	74.3	14.6
Eggs, chicken, fried	72.0	2.4
Chicken, boiled	45.1	3.3
Meat broth	93.8	5.7
Meat, bovine, cooked	55.7	22.8
Meat bovine, fried	48.6	2.47
Meat, lamb, cooked	51.5	0.34
Liver, bovine	71.5	2.42
Fish, River Fried	54.3	17.8
Herring	46.6	0.6
Sardines cans	75.1	0.4
Shrimp	68.0	0.27
Tuna	75.1	0.80
Milk and dairy products		
Milk	88.0	31.5
Yoghurt	56.7	14.0
Cheese, white defatted	70.4	8.2
Cheese, white 20% fat	72.7	45.5
Gouda Cheese	22.1	9.1
Cream	28.0	0.3
Oils + fats	0.0	22.7
Tea and other beverages	98.2	177.2

Upon receipt of all foods indicated in the shopping guide, the raw products were prepared in the total diet laboratory "kitchen" in such a way that only edible parts were used for preparation of diets. Vegetables and fruits were rinsed in cold water as normally in households, and edible parts were taken; then they were combined into main food groups.

The daily intake of 1266.5 g (wet weight) for an adult was used as the basis for calculating the contribution of each of the 76 food components to the diet. A factor was used to calculate the exact quantities of these items needed to prepare 2.5 kg of mixed diet composite. Those foods that were originally in a solid or semisolid state were handled first. Batches of 5 to 10 foods were handled at one time, and weighed portions were pooled in a container and stored frozen until they were blended. The beverages, mainly tea, amounted to more than 10% of the total weight of the diet. The weighed portions were then transferred one by one to a 2 litre glass housing blender fitted with specially fabricated stainless steel blades. The blending procedure was continued until a uniform slurry was obtained, which was freeze-dried. Portioning, weighing and all further handling of the foods were carried out in a class 100 clean room.

The fresh and final weight of the freeze-dried mixed diet were recorded and the percentage moisture was calculated. The freeze-dried samples were ground with titanium knives, and stored in high-density polyethylene bottles 100 ml capacity containing no metal liners that could contaminate the samples, with screw caps. Processing procedure is available in World Health Organization Guidelines (WHO, 1985).

Analytical procedures

The proximate analysis for moisture, protein, fat and ash was done by standard AOAC methods (AOAC, 1990).

Fatty acid determinations

Fat was extracted by blending weighed finely ground samples (2.0 grams) with chloroform-methanol- water (5:10:4 v/v/v) at room temperature. After centrifugation, the chloroform phase was taken to dryness and the total lipid was quantitated gravimetrically. For measurement of fatty acids, a 10 mg aliquot of the extracted lipid was transesterified with boron trifluoride:methanol (Morrison and Smith, 1964). The recovered methyl esters of fatty acids were quantified by gas-liquid chromatography (GLC) with an electron capture detector on a SUPELCO-WAX-10 Supelco (Switzerland) fused silica capillary column 25 m×0.25 mm i.d., as described by Nettleton *et al*., 1990. Fatty acid content is expressed as grams fatty acid per 100 grams total fatty acid.

To convert these values to grams per 100 grams total diet, the total lipid content of the samples was corrected for the presence of non-fatty acid material and the total fatty acid content was multiplied by the percent fatty acid.

Determination of heavy metals

Cadmium (Cd) and lead (Pb) were determined according to Brüggemann and Ocker (1989), using a programmed wet-ashing of the samples. After complexing with sodium diethyldithiocarbarmate and extraction with methyl isobutyl ketone, the minerals were measured using graphite furnace electro thermal atomic absorption spectroscopy (ETAAS).

For mercury, the samples were acid digested (Jorhem *et al*., 1991). Aliquots of the filtrate were transferred with 2.5 ml 2% $SnCl_2$ in 0.7 M H_2SO_4 and the mercury concentration was measured at 240 nm by cold vapour AAS.

Analytical instrumentation

Measurements were carried out on a Perkin Elmer Model 5000 atomic absorption spectrophotometer.

Lead and cadmium were determined by electrothermal atomic absorption spectroscopy (ETAAS) employing pyrolytic plateform graphite tubes and ammonium dihydrogen phosphate (10 g/l) for matrix modification (Brown *et al*., 1988).

Mercury was analyzed by the 'cold vapour' technique (CVAAS) by treatment with $NaBH_4$ to give Hg vapour.

Instrumental parameters

	Cd	Pb	Hg
Wavelength (nm):	228.8	217	240
Detection limits (μg/kg)	5	10	5

Quality control

With each analytical batch, the series included analysis of 3 reagent blanks to monitor contamination and estimate detection limits, at least one spiked recovery and at least one standard reference material (SRM), Standard reference materials (SRM) were analyzed to check the accuracy and precision of the methods and were obtained from the National Bureau of Standard RM 8431 (Mixed diet) containing 42, 155 and 7.4 μg per kg of Cd, Pb and Hg, respectively.

Calculation

The mean absorbance produced by the standards (corrected for the standard blank) was plotted vs the concentration of the analyte in the sample to produce an external calibration curve. The concentration of the analyte in the sample was calculated from the following equation:

$$[M] = (C \times V)/SW$$

where $[M]$ is the concentration (ng/ml) of Cd; Pb or Hg in original sample, C is the the respective concentration in the analytical sample as calculated from a standard curve in units of ng/ml, V is the volume of the analytical sample in units of ml, and SW is the weight of the sample employed in units of grams. The mean (plus or minus one standard deviation from the mean) were determined per 100 g sample.

Determination of nitrates (British Standards Institution, 1976)

The method was based on the extraction of the sample with hot water, precipitation of the proteins with zinc acetate and reduction of the extracted nitrate in the clear filtrate to nitrite by metallic cadmium. After development of a red colour by addition of sulphanilamide and N-l-naphthylethylenediamine dihydrochloride to the filtrate, photometric measurement was carried out at a wavelength of 538 nm. Standard potassium nitrate solution was used as the external calibration curve.

Chlorinated pesticide and polychlorinated biphenyls (PCB)

The methods used were Soxhlet extraction of lipophilic hydrocarbons and separation of the coextracted fats onto basic aluminium oxide according to Steinwandter and Buss (1975). The eluates were further purified on a mini-silica gel column (Specht and Tillkes, 1980). Throughout all preparation and analysis steps, recovery was determined as reported by Brunn *et al.* (1989) and Georgii *et al.* (1994). The detection limit for the method described above was 1μg, (PCB 28)/kg fat.

The standard OCPs and PCBs (nomenclature of PCB congeners according to Ballschmiter and Zell, 1980) used were: Aldrin; Bromocyclen; Dieldrin; Endrin; Ketoendrin; Hexachlorobenzene (C_6Cl_6), α-, β-Hexachlorocyclohexane ($C_6H_6Cl_6$), γ-Hexachlorocyclohexane (γ-HCH Lindane^R), p,p′-DDT(2,2-bis(p-chlorophenyl)-1,1,1-trichloroethene). o,p′-DDT, p,p′-DDE (2,2-bis (p-chlorophenyl)-1,1-dichloroethene), o,p′-DDE, p,p′-DDD (2,2-bis(p-chlorophenyl)-1,1-dichloroethene), and o,p′-DDD.

Heptachlor, cis-and trans-Heptachloroepoxide, oxychlordane, cis- and trans-chlordane, α-endosulfan, β-endosulfan, and endosulfansulfate.

PCB 28 (2,4,4′-tri), 31 (2,4′,5-tri), 49 (2,2′,4,5′-tetra), 52 (2,2′,5,5′-tetra), 101 (2,2′,4,5,5′-penta), 138 (2,2′,3,4,4′,5′-hexa), 153 (2,2′,4,4′,5,5′-hexa), 156 (2,3,3′,4,4′,5-hexa), 180 (2,2′,3,4,4′,5,5′,-hepta), 183 (2,2′,3,4,4′,5′,6-hepta), and 189 (2,3,3′,4,4′,5,5′-heptachlorobiphenyl) (all standards supplied from Promochem GmbH, Wesel, Germany)

Apparatus and analytical conditions

Two Hewlett Packard instruments Model 5890/2 and 5890/3 gas chromatographs (GC) with electron-capture detector were employed for the determinations. Two 60 m fused silica capillary columns were used (DB 1 and DB 5 columns from J&W/Fisions, Mainz, Germany, of 0.25 mm inside diameter, 0.25 μm film thickness, with carrier gas hydrogen, splitless injection, and a linear temperature-programming (according to Georgii *et al.*, 1994). The specificity of the gas chromatographic analysis was checked by mass spectrometry. Recovery and accuracy were also confirmed (Georgii *et al.*, 1994). by analysis of certified butterfat with known PCB concentrations (Promochem GmbH, Wesel, Germany).

Assessments of dietary nitrate, pesticides and PCB intakes. Mean composition data analyzed in the present work were combined with consumption data obtained from an earlier food consumption Survey on a subpopulation group (Hussein *et al.*, 1995) to compile and estimate daily intakes.

RESULTS

The amounts of food commodities used for the preparation of the mixed diet are shown in Table 1.

Table 2 presents the proximate composition of the total diet. Fat makes up on the average 14.12% of the dry matter.

The fatty acid compositions of the mixed diets are shown in Table 3. Saturated fatty acids made up 41.1–51.5% of all fatty acids, Palmitic acid (C16:0) was the major saturated fatty acid, comprising 29.6–32.0% of fatty acids, and accounting for 60.1–89.9 percent of all saturates, followed by stearic acid (C18:0) with values ranging from 6.9 to 11.6% of fatty acids.

The majority of fatty acids, about 31.7–38.9%, are monounsaturates with oleic acid (C18:1) predominating in this class and comprising 31.0–34.9% of fatty acids.

Linoleic acid (C18:2) is the predominant polyunsaturated fatty acid in the Egyptian total diets accounting for 92.8–95.8% of all polyunsaturates with values ranging from 13.5 to 25.3 of total fatty acids.

Total omega-3 fatty acids (C18:3, C20:5, C22:6) account for 4.2–7.1% of the polyunsaturates. Linolenic acid (C18:3–n3) content ranged from 0.62–1.89% of total fatty acids, accounting for 72–100% of total omega-3 fatty acids in the mixed diet.

The long-chain omega-3, 20:5 and 22:6 in the Egyptian total diet, are present in traces.

Table 4 presents heavy metals, nitrate and nitrite and organochlorine concentrations in the total mixed diets. Intake values were estimated and compared with the provisional tolerable weekly intakes (PTWI) (Codex Alimentarius Commission, 1984).

DISCUSSION

The amount of lipid consumed by the subjects in the present study ranged from 16–43 g/day and averaged 24 g/day per person. This mean value is much lower than the estimate of total fat intake in European food with 73 g for South Italy and 122 g for Germany (Cresta *et al.*, 1969), and our mean figure is much lower than the estimate of 169 g/day per capita in the United States (Rizek *et al.*, 1983).

In terms of foods, the Egyptian diet is defined as being a diet high in cereals (more than 60 percent of total energy) and low in total fats (less than 30% of total energy), with moderate amounts of added fats. The mean M/S ratio of the mixed diets is 0.78 ± 0.04. This is similar to the M/S ratios (around 0.8) in the

Table 2. The proximate composition of the total diet g/100 g fresh weight

Composite	Mean	± SE
Moisture	63.05	0.107
Protein	5.0	0.029
Fat	5.5	0.424
Ash	1.3	0.005
Carbohydrate	21.1	
Calories (kcal)	142	

Table 3. Fatty acid composition of lipids from composite total diets (g/100 g total fatty acids)

Fatty acid	Mean	SE	Range
Saturated			
12:0	1.67	0.388	0.67–2.32
14:0	4.33	1.64	0.89–8.23
16:0	31.0	0.517	29.6–32.0
17:0			
18:0	9.86	1.045	6.89–11.62
20:0	0.10	0.103	0.0–0.41
Total (S)	47	2.24	41.1–51.5
Monoenes			
14:1	1.06	0.793	0.06–3.41
16:1	1.25	0.241	0.62–1.71
18:1	32.71	1.007	31.00–34.9
20:1	0.0	0.0	0.0–0.0
Total (MUFA)	35.2	1.55	31.7–38.9
Dienes			
18:2	16.80	2.848	13.52–25.33
20:2	0.0	0.0	0.0–0.0
Trienes			
18:3—*n* 6	0.013	0.013	0.0–0.05
18:3—*n* 3	0.983	0.303	0.62–1.89
20:3—*n* 6	0.0	0.0	0.0
20:3—*n* 3	0.0	0.0	0.0
Tetraenes			
18:4	0.013	0.013	0.0–0.05
20:4	0.0	0.0	0.0–0.0
20:5	0.04	0.040	0.0–0.16
22:6	0.02	0.020	0.0–0.08
n 3—FA	1.05	0.290	0.62–1.89
n 6—FA	16.8	2.84	13.6–25.3
n 6—n 3-ratio	17.6	2.26	13.4–23.00
MUFA	35.2	1.55	31.7–38.9
SFA	4.30	1.96	40.4–49.2
PUFA/SFA ratio	0.404	0.090	0.294–0.673

German (0.77) and Northern Italian (0.67) diets, except for a value of 1.5 in Holland and for the remarkable figure of 3.9 obtained in Southern Italy (Ferro-Luzzi and Sette, 1989).

Table 4. Toxic residue contents in the mixed diet and estimated daily intake

Toxic substance	Unit	Concentration per kg dry matter	Concentration per kg diet	Daily intake	PTWI*
Cadmium	μg	39.5	15.5	19.7	70
Lead	μg	485	191	242	430
Mercury	μg	158	62.0	78.5	43
$NO_2 + NO_3$	mg	593	233	296	
Males	mg			255	
Females	mg			211	
Pesticides					
Lindane	μg	169	9.38	119	
p.p. DDE	μg	30.9	1.72	2.18	

FAO/WHO provisional tolerable weekly intakes converted to represent daily intake (Codex Alimentarius Commission, 1984).

The mean P/S ratio was 0.404 ± 0.09; this figure was slightly lower than the P/S ratios of the separated fats in Southern Italy (P/S ratio = 0.53) (Ferro-Luzzi and Sette, 1989).

It is now accepted that the dietary fatty acid pattern of total diets reflects their food sources. Saturated fatty acids, typified by stearic and palmitic, are found mostly in animal tissue. Oleic acid (cis-6-octadecanoic acid) is abundant in vegetable oil. According to Mead (1985) oleate is not a good PUFA precursor since its further desaturation is largely suppressed.

Linoleic acid, an essential fatty acid is an omega-6 fatty acid; i.e. the first double bond from the omega carbon is between the sixth and the seventh carbon atoms.

Adequate linolenic acid is reported to fluctuate from 0.5–1.0% of the total dietary energy (Mclean and Sinclair, 1985). In the present mixed diets, linolenic acid made up 0.17–0.68 with a mean figure of 0.35 ± 0.134 of the total dietary energy, suggesting poor linolenic acid status.

Eicosopentenoic acid (20:5) and docosahexenoic acid (22:6) are the most abundant long-chain omega-3 fatty acids found in the triglycerides of phytoplankton and are therefore identified in highest concentrations in ocean fish, salmon and mackerel (Weaver and Holob, 1988).

Polyunsaturated fat available in 20 Western countries, is reported to range from 5 g/person in Finland up to 26 g/German adult/day (Wahlquist, 1985). Based on the present results, an intake estimate of 12.4 g PUFA was consumed by an Egyptian adult per day.

The ratio of intakes of polyunsaturated to saturated fatty acids or the P/S ratio, is considered a measure of the atherogenicity of the diet; i.e. the lower the estimated ratio, the more atherogenic is the diet.

In the USA, dietary goals have been formulated (Gordon *et al.*, 1985) recommending that 30% of the energy be derived from fat, with equal contribution from PUFA, SFA and MUFA, a P/S ratio of 1.0, and 300 mg of cholesterol per day.

Chemical residues and contaminants

Average total dietary cadmium intake (19.7 μg/d) was about 28% of the PTWI. This level is slightly higher than figures reported for Germany (14.3 μg/d) (Stelz *et al.*, 1990) and those reported in Spain (16 μg/d) (Kumpulainen, 1996).

Dietary lead intake amounted to 242 μg/day, representing 56.2% of the PTWI as established by the FAO/WHO for this metal. This value is much higher than those reported in the Netherlands (32 μg/d) (de Vos *et al.*, 1984), the USA (11.6 μg/d) (Capar, 1990) and Germany (77.7 μg/d) (Stelz *et al.*, 1990).

The highest lead dietary intake (179 μg/d) have been reported in Belgium (Buch *et al.*, 1983), Italy (280 μg/d) (Kumpulainen, 1996).

The average daily intake of mercury (76.4 μg/d) is quite high and about 177% of the PTWI established by the FAO/WHO (Codex Alimentarius Commission, 1984). Fishes and other seafoods have been reported to be the major Hg contaminants (60–90%). It is notable that fishes did not contribute significantly to the mixed diet composite. Poultry has been reported to contribute to 27% to the mercury intake (Kumpulainen, 1996).

The dietary high lead and mercury intakes would be of concern, particularly in terms of the risk of detrimental health effects for children and pregnant and lactating women. This contamination could be occasional. Further studies are needed in order to more closely assess the mercury and lead intakes as well as identify mercury and lead sources.

Based on the present analytical data of mixed diet composite, dietary ingested nitrate and nitrites amount to 296 mg/day. This figure approaches 140% of the acceptable daily intake with respect to females (211 mg/day) and is roughly 116% of the acceptable daily intake with regard to males (255 mg/d) (Bundesrats-Drucksache No. 273, 1994).

The average western diet was found to contain 62–124 mg nitrate/person/day (Hotchkiss, 1988) and the dietary intake of nitrate can increase substantially by consuming vegetables and/or high nitrate water (Hotchkiss *et al*., 1992).

Due to a widespread concern over environmental contamination by the chlorinated hydrocarbon insecticides, the usage of such compounds has been dwindling rapidly in recent years. Intakes of lindane^{R}, dieldrin, endrin and DDT, with the daily Egyptian diet in 1989, have been calculated to be: 702, 95, 167 and 958 μg, respectively per subject per day (Abdel-Gawaad and Shams El-Dine, 1989). Respective acceptable daily intakes were 700, 7, 14 and 1400 (Codex Alimentarius Commission, 1984).

The results presented in Table 4 show much lower mean figures of 11.9 μg for γ-HCH (lindane^{R}) than the figure reported eight years ago (702 μg). DDT was completely absent in the composite diet and only the degradation product p,p′-DDE is present. The results showed that polychlorinated biphenyls (PCBs) and the other organochlorines investigated were below the limit of detection.

The present finding demonstrates that there is a clear trend towards lowering pesticide residues in food commodities, in the line with the country's policy.

CONCLUSION

The consultation of the European cooperative network on trace elements highly recommended that sample collection should be carried out over a period of at least three years for estimating the annual variation in heavy metal contamination of staple foods. Such a plan of work awaits further investigation.

ACKNOWLEDGEMENTS

One of the authors (Z.A.S.) was a recipient of a grant from the Deutsche Forschungsgemeindschaft in Bonn-Bad-Godesberg, Germany. The authors acknowledge the excellent cooperation of Professor Dr H. Daniel, Professor Dr E. Muskat and Dr A Stelz. The authors acknowledge the technical help of Mrs B. Hecker, Mr Peter, Mrs A. Omar and Mrs R. Schwirz.

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