AMYLASE AND LIPASE VALUES IN NORMAL SUBJECTS

H. G. VAN RIET AND J. O. O. HOEKE

Departments of Gastroenterology and Clinical Chemistry**, University Medical Clinic***, Utrecht (The Netherlands)*

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SUMMARY

In 146 hospitalized individuals (71 men and 75 women), the simultaneous fasting serum amylase and lipase concentrations and the urinary amylase excretion per 24 h were determined. Patients with pancreatic abnormalities or other diseases which might produce abnormal enzyme values were ruled out from this study.

Age and sex were found not to influence the parameters under investigation. The various parameters failed to show a distinct correlation. The renal amylase clearance was calculated for the individuals, and the frequency distribution is presented. In 10 normal subjects, the postprandial 3-h urinary amylase excretion was found not to differ from the preprandial value. In 10 normal subjects, the course of the 24-h urinary amylase excretion showed no distinct rhythm. The urinary amylase excretion per 24 h probably constitutes a more reliable parameter than the excretion during a brief period of collection or the fasting amylase concentration.

The clinical usefulness of some chemical parameters of pancreatic function – urinary amylase, serum amylase and serum lipase – requires knowledge of their normal ranges, preferably determined in the same laboratory. In order to establish these ranges, a study was made in a number of hospitalized patients. Data collected on urinary amylase concerned not only the concentration in a fasting urine sample but also the urinary excretion of this enzyme per 24 h. Gambill and Mason[1] pointed out that the urinary amylase excretion per unit of time should supply more clinical information than the amylase concentration in fasting urine.

Normal values determined in this manner would not *per se* warrant publication. However, the data collected supplied information also about: a. The influence of age and sex. b. The interrelations between the various parameters. c. The renal amylase clearance. d. The changes in urinary amylase output, both postprandial and in the course of a 24-h period.

* Head: O. J. ten Thije M. D.
** Head: J. de Wael Ph. D.
*** Heads: Prof. F. L. J. Jordan M.D. and Prof. L. A. Hulst M.D.

SUBJECTS AND METHODS

From 146 patients (71 men and 75 women) hospitalized in the Medical Department of the Utrecht University Hospital, without any known abnormalities of the abdominal organs or salivary glands, or vague abdominal symptoms of obscure aetiology, fasting urine was collected for determination of the amylase concentration. This was immediately followed by venipuncture for determination of serum amylase and lipase concentrations. Subsequently, urine was collected over a 24-h period for the determination of amylase output.

The patients were divided into 10-year age groups, the first age group being 10–20, the seventh and last group including the patients aged 70–90. The number of patients in each group averaged more than 10 (range: 8–15).

Moreover, in a group of 14 healthy nurses, technicians and physicians, the following study was made.

1. In order to establish the influence of a meal on urinary amylase excretion, 10 of these test subjects partook of a hospital meal of bean soup, sauerkraut with sausage, and fruit custard. Urine was collected during the last 3 preprandial hours and the first postprandial one, and the amylase concentration was determined.

2. In order to establish variations in the amylase excretion, 10 of these 14 individuals supplied 24-h urine, collected in 3-h portions. During this 24-h period of collection these subjects adhered to their normal activities, eating habits, etc., as far as possible.

Amylase assays were carried out as described by Street and Close [2,3], with the following modifications:

(1) the concentration of the amylose solution was increased from 1.0 g/l to 1.5 g/l in order to ensure a sufficient substrate excess during the period of incubation;
(2) the volume of 0.01 *N* iodine solution added after incubation was increased from 0.6 ml to 0.7 ml to overcome interference by serum proteins[4];
(3) when the absorbance value obtained appeared to be less than 50% of the unincubated control, determination was repeated after appropriate dilution.

The results are presented in micromoles of reducing groups formed per min by 1 l of serum or urine (units recommended by the Commission on Enzymes of the International Union of Biochemistry). For this purpose a number of results obtained by the above mentioned method were compared with those obtained when the increase in concentration of reducing groups in our incubation mixture was determined by the method of Nelson–Somogyi[5].

The results were calculated using the formula:

$$\frac{A_{\text{control}} - A_{\text{test}}}{A_{\text{control}}} \times F_{\text{dil}} \times C$$

where

A_{control} = Absorbance at 657 nm of an unincubated mixture of all reagents and the sample;
A_{test} = Absorbance at 657 nm obtained when the determination was carried out including the incubation at 37°;
F_{dil} = Dilution factor;

C = Conversion factor obtained by comparison with the results of the Nelson–Somogyi method.

Lipase determinations were made according to Tietz *et al.*[6]. Titration results were re-calculated to give the number of micro-equivalents of acid liberated per min by 1 l of serum.

RESULTS

No significant difference between the sexes was demonstrable in any of the parameters studied (urinary amylase concentration, urinary amylase excretion per 24 h, serum amylase and lipase concentrations). Fig. 1 shows that no distinct influence of

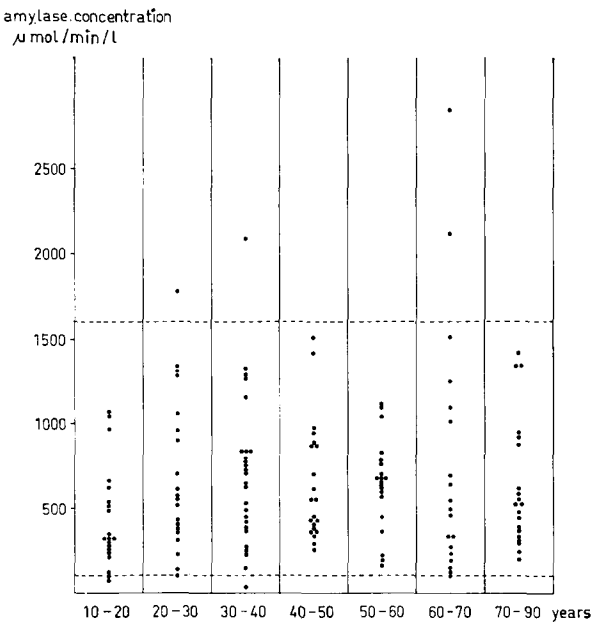

Fig. 1. Age distribution of urinary amylase concentration in 146 fasting persons.

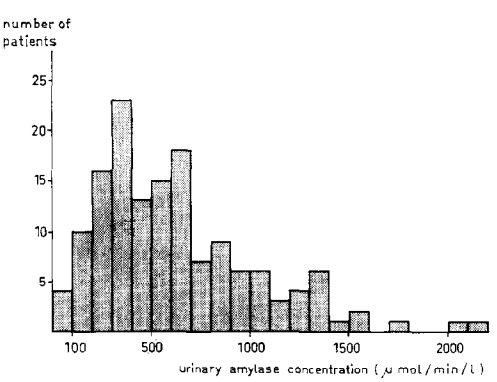

Fig. 2. Frequency distribution of urinary amylase concentration in 146 fasting persons.

the test subjects' age on the urinary amylase concentration was observed. Analogous results were obtained for the other parameters (total urinary amylase excretion per 24 h, serum amylase and lipase concentrations).

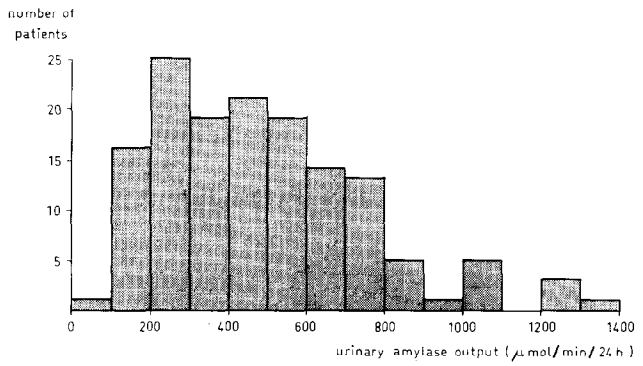

Fig. 3. Frequency distribution of urinary amylase output per 24 h in 144 persons.

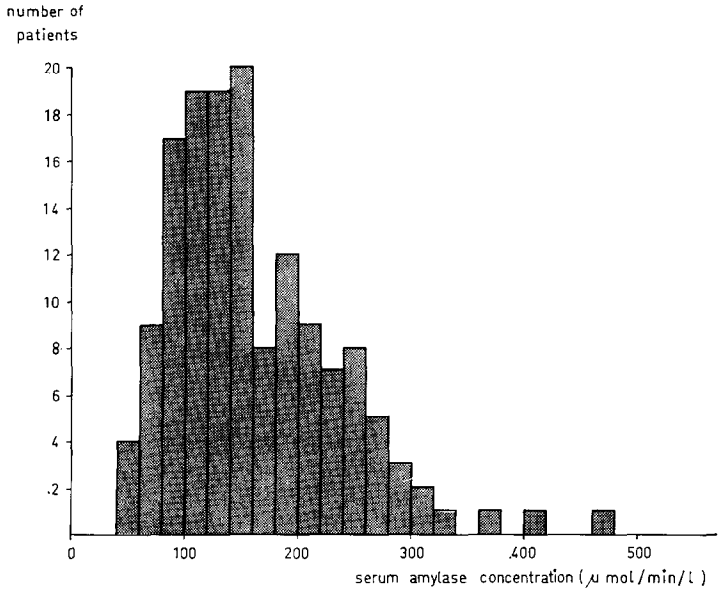

Fig. 4. Frequency distribution of serum amylase concentration in 145 persons.

In view of the above, for each of the parameters all data were combined regardless of age and sex, and frequency distributions were plotted in block diagrams. Figs. 2, 3, 4, and 5 thus present the data on amylase concentrations in fasting urine, total urinary amylase excretion per 24 h, serum amylase and serum lipase concentrations respectively. Since no Gaussian curve was obtained in any of the cases, the limits were determined exclusively by graphic analysis of the data; the limits were chosen so as to encompass 95% of the observations.

	Lower limit	*Upper limit*
Amylase concentration in fasting urine	100 μmol/min/l	1600 μmol/min/l
Total amylase excretion per 24 hours	120 μmol/min/l	1200 μmol/min/l
Amylase concentration in serum	60 μmol/min/l	300 μmol/min/l
Lipase concentration in serum	0 μeq/min/l	80 μeq/min/l

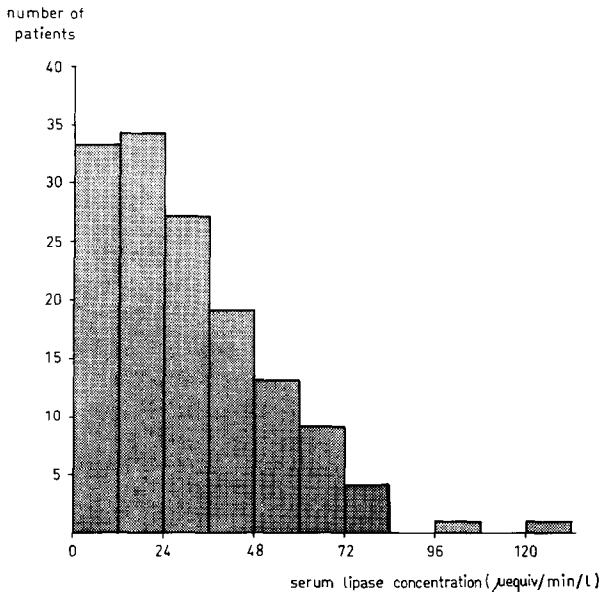

Fig. 5. Frequency distribution of serum lipase concentration in 145 persons.

There is no distinct correlation between the simultaneously determined urinary amylase excretion per 24 h and serum amylase concentration (Fig. 6). It will be obvious that the amylase clearance, too, can be calculated from the data obtained (Fig. 7). The limits of clearance (determined as mentioned above) were 0.6–4.2 ml/min.

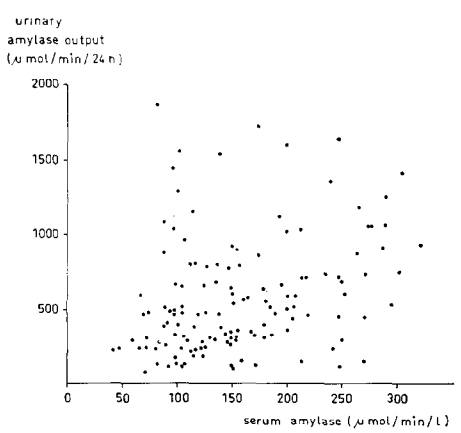

Fig. 6. Urinary amylase output per 24 h *vs.* serum amylase concentration in 144 persons.

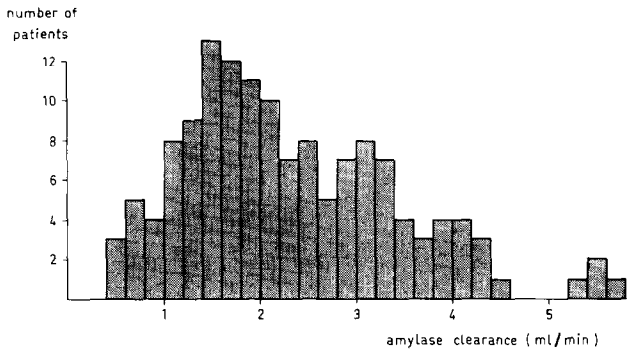

Fig. 7. Frequency distribution of amylase clearance in 144 persons.

A scatter diagram of individual serum amylase over serum lipase concentrations is presented in Fig. 8. A conspicuous feature is that lipase concentration was often not measurable in serum from normal subjects. Therefore the line which might be constructed in the dot diagram very clearly fails to pass through the zeropoint.

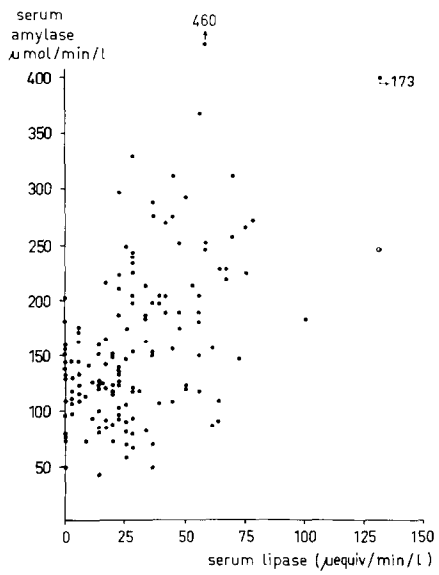

Fig. 8. Serum amylase concentration *vs.* serum lipase concentration in 145 persons.

Fig. 9 shows the course of urinary amylase excretion per 24 h in 10 individuals. The excretion, measured in 3-h portions, showed no distinct rhythm. Theoretically, small postprandial peaks in output might have been missed but, when 24-h urine was collected in 1-h portions (in one case), no postprandial peaks were observed either.

Fig. 10 indicates that the preprandial and postprandial urinary amylase concentrations in 10 normal test subjects varied widely. However, the amylase excretion per 3-h portion of urine hardly changed after ingestion of a hospital dinner in these subjects.

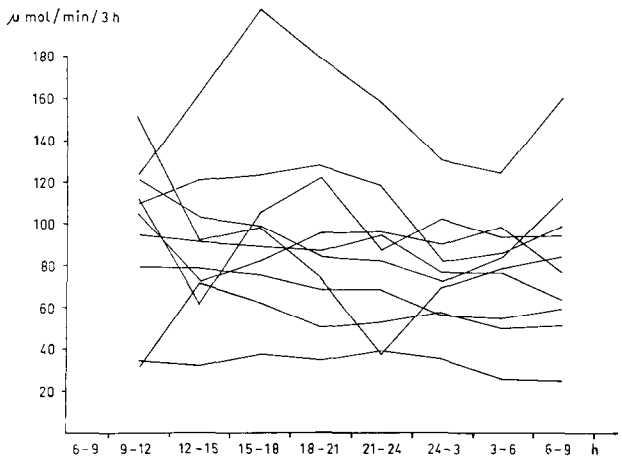

Fig. 9. Urinary amylase output (3-h portions) in the course of 24 h in 10 normal persons.

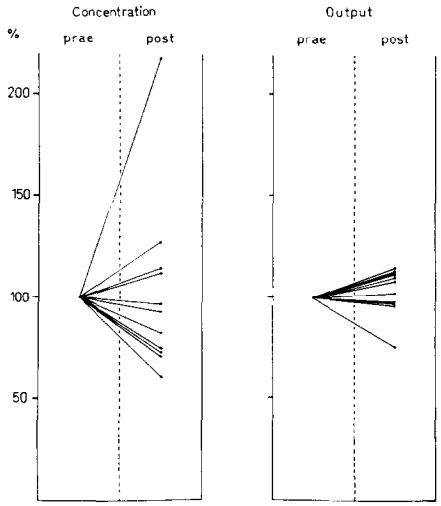

Fig. 10. Postprandial urinary amylase concentration and output in 10 normal persons in percent of the preprandial value.

DISCUSSION

As we pointed out, modifications of the methods of determination described in the literature were used in the study under discussion. This is the principal reason why our numerical values cannot be compared with those obtained by other investigators. Nevertheless, the conclusions which can be drawn retain their general validity. The distribution curves obtained are asymmetrical. In contrast to this, Berk *et al.*[7] found a typical Gaussian distribution of serum amylase concentrations in 67 normal subjects (medical students); for the serum lipase concentrations, however, these authors did find an asymmetrical distribution. The question arises whether the asymmetry of curves in the present study does not, after all, reflect the composition of the group

studied. We deliberately chose hospitalized individuals for this purpose for, if enzyme determinations are to be evaluated in clinical patients, then one must dispose of "normal" values concerning individuals under otherwise entirely identical conditions. Although the group was selected so as to rule out pancreatic pathology or other known causes of increased enzyme activity, it must nevertheless be borne in mind that the individuals selected were patients. Perhaps, causes of increased activity of the enzymes studied are more prevalent in hospitalized patients than in a healthy, youthful control group – even though patients with known pancreatic conditions, etc., are ruled out.

Individuals whose amylase and lipase concentrations exceeded the 95% interval, showed no common clinical characteristic that might indicate a correlation between the clinical picture and these high values. The fact that serum lipase values were not measurable in 14 individuals must probably be ascribed to lack of sensitivity of the method of determination, which produces inaccurate results at low lipase concentrations.

The question whether the accepted lower limits – which generally receive little attention – constitute useful clinical parameters cannot be answered until pathological sera and urines are included in the investigation. The same applies to the question whether urinary amylase excretion is clinically more valuable than amylase concentration in fasting urine.

The lack of a distinct correlation between serum amylase and lipase levels, and between serum amylase concentration and urinary excretion, is a conspicuous finding. In the study of urinary amylase excretion per unit of time it was interesting to observe fluctuations in the course of a 24-h period, particularly after meals. The question arises whether in patients with pancreatic affections there was a more than normal effusion of amylase into the circulation and increased urinary excretion after meals ("Ernährungsbedingte Fermententgleisung"[8]).

Fig. 9 shows that in 10 normal test subjects excretion during a 24-h period takes an unpredictably irregular course, showing no systematic diurnal peak, nor a nocturnal dip, which is in agreement with recent findings by Myhre *et al.*[9]. In view of the irregular excretion we believe that collection of urine during a 24-h period should be preferred to collection during a shorter period (2 h) as recommended by Saxon *et al.*[10].

Reduction of the urine output as a result of some renal disease may be accompanied by an increase in serum amylase concentration. In view of the amylase clearance found, the increase per day can be estimated as maximum 10–20% of the normal serum amylase concentration; this may make the differential diagnosis from pancreatic affections difficult. Attempts to increase the diagnostic value of amylase determination by investigating isoamylase activities have been reported by several investigators[11,12]. However, Wilding[4] and Hoeke *et al.*[13] demonstrated that the proposed methods appear to lead to erroneous interpretations. In these cases it is therefore preferable to determine the serum lipase concentration as well[14].

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