DOI: 10.4149/BLL_2016_125

Bratisl Med J 2016; 117 (11)
648 – 652

EXPERIMENTAL STUDY

The value of prolidase enzyme in rats with experimentally induced mild and severe pancreatitis

Yalcin M[1], Oter V[2], Ozgonul A[3]

Department of General Surgery, Mehmet Akif Inan Teaching and Research Hospital, Sanliurfa, Turkey.
drmetinyalcin@hotmail.com

ABSTRACT
INTRODUCTION: Acute pancreatitis is basically considered as activation of inactive proenzymes in the pancreas and digestion of the gland itself. This study was performed to determine if prolidase enzyme, which plays a role in collagen metabolism, could be used as a parameter to assess the severity of pancreatitis in experimentally induced mild and severe pancreatitis.
MATERIAL AND METHOD: To create experimentally induced acute pancreatitis 0.1 ml of normal saline solution (NSS) was given five times with an interval of one hour to rats in the first group; 50 µg/kg of cerulein five times with an interval of one hour in the second group; 80 µg/kg of cerulein five times with an interval of one hour in the third group, in the form of intraperitoneal injection.
RESULTS: When the serum prolidase values at beginning, 1st, 5th and 24th hours in group II and III were compared among themselves, there was a statistically significant increase($p < 0.05$). The evaluation between groups revealed a statistically significant increase in the value of serum prolidase in group II and group III compared with the control group ($p < 0.05$). In comparisons performed with tissue values, a statistically significant increase determined in the value of serum prolidase in group II and group III compared with the control group was observed ($p < 0.05$).
CONCLUSION: The findings obtained in our study showed that prolidase activity increases directly proportionaly with the severity of pancreatitis. This allows us to postulate that prolidase enzyme activities provide guidance about the metabolism of collagen in patients with acute pancreatitis, serious damage occurring in collagen protein and metabolic control is further distorted depending on the duration and intensity of damage but to be able to speak more precisely, there is a need for further, more detailed and extensive researchs *(Tab. 8, Fig. 2, Ref. 30)*. Text in PDF *www.elis.sk.*
KEY WORDS: prolidase enzyme, rat, acute pancreatitis, pancreas, enzyme activity.

Introductıon

Although acute pancreatitis is a disease known since ancient times, first description was made by Chiari in 1883, and the first classification was made by Fitz in 1889 (1). Acute pancreatitis is basically considered as activation of inactive proenzymes in the pancreas and digestion of the gland itself (2).

Although, over the past decade, acute pancreatitis created in experimental animal studies not fully replicates acut pancreatitis in humans, it is known that intracellular early structural and biochemical changes exhibit similarities but significant progress in the treatment field has not been achieved.

Collagen, forming the basic structure of connective tissue skeleton, plays a key role in inflammation and wound healing and is the most abundant protein in the body. Collagen can be affected by pathological events of many organs, tissues and cells because it is the structural component of extracellular matrix, beside many organs and tissues (3).

Prolidase enzyme is a metalloenzyme that belongs to the hydrolase family and is activated by mangan (Mn^{2+}). Prolidase has great importance in the collagen cycle. Because prolidase enzyme is the only enzyme which catalyzes intracellular rapid hydrolysis of compounds having a peptide bond containing an imino nitrogen of proline and hydroxyproline that occurs in the final step of collagen catabolism (X-proline or X-hydroxyproline), metabolism of collagen could be determined by looking at the prolidase enzyme activity in the blood and tissue levels (4, 5).

Extensive tissue distribution of collagen suggests that changes in prolidase enzyme activity, which is a spesific enzyme for its turnover, may be important for development of many diseases. In a few studies that evaluate prolidase enzyme activity in diseases that progress with inflammation, prolidase enzyme activity was significantly higher due to collagen destruction (6).

This study was performed to determine if prolidase enzyme, which plays a role in collagen metabolism, could be used as a parameter to assess the severity of pancreatitis in experimentally induced mild and severe pancreatitis.

[1]Department of General Surgery, Mehmet Akif Inan Teaching and Research Hospital, Sanliurfa, Turkey, [2]Department of Gastroenterological Surgery, Mehmet Akif Inan Teaching and Research Hospital, Sanliurfa, Turkey, and [3]Department of General Surgery, Medicine Faculty, Harran University, Sanliurfa, Turkey

Address for correspondence: V. Oter, Department of Gastroenterological Surgery, Mehmet Akif Inan Teaching and Research Hospital, Esentepe Quarter, Ertugrul Street, Sanliurfa, Turkey.
Phone: +5053366333, Fax: 04143186707

Acknowledgement: This study was performed in Harran University Faculty of Medicine, Laboratory of Animal Experiments, after approval of the Ethics Committee (issue number 270-25 and decree number 2009/08). The study was supported by Harran University Coordination of Scientific Research Projects (HÜBAK-Project No. 938).

Material and method

This study was performed in Harran University, Faculty of Medicine, Laboratory of Animal Experiments, after approval of the Ethics Committee (issue number 270-25 and decree number 2009/08). The study was supported by Harran University Coordination of Scientific Research Projects (HÜBAK-Project No. 938).

3–3.5 months old, weights ranging between 140–150 g, 30 Wistar albino female rats were used in the study.

Three groups, 10 rats in each group, were created for the study. Group 1 served as control group, Group 2 as mild pancreatitis group, and Group 3 as severe pancreatitis group.

To create experimentally induced acute pancreatitis 0.1 ml of normal saline solution (NSS) was given five times with an interval of one hour to rats in first group; 50 µg/kg of cerulein, five times with an interval of one hour in second group; 80 µg/kg of cerulein, five times with an interval of one hour in third group, in the form of intraperitoneal (ip) injection.

Just before the injection of cerulein and 1, 5 and 24 hours after the cerulein injection, tail blood taken from all the rats and serums obtained were analyzed biochemically. Serum amylase, lipase, AST, ALT, WBC, LDH, glucose, total bilirubin, direct bilirubin, GGT and ALP activity were tested with commercially obtained kits (Boehringer Mannheim, Germany) and biochemical pancreatitis table observed.

Blood samples obtained from rats were stored in heparinized biochemical tubes and serum samples separated after 10 minutes of centrifugation under 3000 rpm. Serum samples were stored in –80 °C until the day that prolidase enzyme analysis was performed. Serum samples were dissolved at analysis day and serum prolidase enzyme analysis performed spectrophotometrically by Chinard method which is a colorimetric assay method. After 24 hours, abdomens of all the rats were opened after decapitation. Pancreatic tissue was completely removed. Pancreas tissue was divided into two portions and first part kept frozen for biochemical analysis and other part kept for histopathological examination. Specimen kept for histopathological examination was stored in 10 % formaldehyde solution and sent to the pathology laboratory for further evaluation. Sections four micrometer in size were cut from each pancreatic tissue and stained with hematoxylin-eosin. Pancreatic tissues were examined under a light microscope.

Histopathological examination was performed by Schoenbergs' pancreatic stage and grade scale (Tab. 1) (7, 8). Pancreatic tissues were examined by a pathologist who is an expert in groups under light microscope.

According to this, histopathological edema, neutrophil infiltration and cell damage were evaluated (Tab. 2).

Tab. 1. Schoenberg's pancreas grade and stage scale.

Assessing Edema	
0	No edematous tissue
1	Intralobular edema
2	Moderate interlobular and intraacinar edema
3	Severe interlobular and intraacinar edema
Assessing Inflammation	
1	Presence of neutrophils in the border of capillaries and post-capillary venules
2	Presence of neutrophils in perivascular tissue
3	Diffuse infiltration surrounding the pancreatic gland
Assessing Cell Damage	
1	1–4 necrotic cells
2	5–10 necrotic cells
3	11–16 necrotic cells
4	>16 necrotic cells

Tab. 2. Histopathological pancreatitis scoring.

Score between 2–4	Mild pancreatitis
Score between 5–8	Moderate pancreatitis
Score over 8	Severe pancreatitis

Statistical analysis

Statistical analysis of blood and tissue samples was accomplished using the SPSS program for Windows 16.0 (Statistical Package for Social Sciences for Windows 16, inc, USA). Nonparametric tests like Mann–Whitney U test, Kruskal–Wallis analysis, Repeated Variance analysis and Kolmogorov–Smirnov test were used for the comparison of data. The results are presented as mean ± standard deviation and Repeated management. Serum prolidase level was calculated using Student's t-test. A p value under 0.05 was considered to be significant.

Results

Biochemical evaluation

After the analysis of serum biochemical parameters between groups at beginnig, at 1st, 5th and 24th hour, amylase and lipase levels were higher in group II (mild pancreatitis) and group III (severe pancreatitis) compared with the control group and this result was statistically significant ($p < 0.05$), (Fig. 1). Other serum parameters; AST, ALT, WBC, LDH, glucose, total bilirubin, direct bilirubin, GGT and ALP levels have revealed no statistically significant difference between groups ($p > 0.05$).

Serum and tissue prolidase levels at the beginning, 1st, 5th and 24th hour were compared between groups and in each group

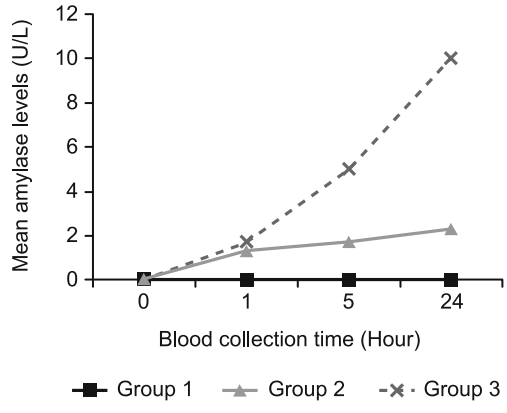

Fig. 1. Amylase levels in groups at hours 0, 1, 5 and 24.

Tab. 3. Group I (control) evaluation of serum biochemical values.

Variables	0th hour	1st hour	5th hour	24th hour	P value
Prolidase	752±10.4	751±13.4	752±14.3	750±9.59	0.725
Glucose	1.34±0.34	1±0.20	20±6.98^{bd}	52±13^{cef}	< 0.001
WBC	2.71±0.94	4.04±1.30	2.98±0.85	4.32±1.20^{c}	0.023
AST	94±25	90±14	107±26	99±22	0.336
ALT	41±14	33±10	50±16	41±7.93	0.278
GGT	2.68±0.89	3.43±1.12	3.41±1.31	4.19±1.66	0.039
ALP	47±10	55±5.55	57±11^{b}	66±14^{c}	< 0.001
T. bil	0.48±0.12	0.65±0.18	0.10±0.04^{bd}	0.08±0.02^{ce}	< 0.001
D. bil	0.17±0.49	0.16±0.03	0.05±0.01^{bd}	0.03±0.01^{ce}	< 0.001
Amylase	60±17	60±12	62±16	69±17	0.186
Lipase	6.68±1.73	7.03±1.54	6.11±0.80	6.94±0.80	0.940

[a]: There is a significant difference between 0 and 1, [b]: There is a significant difference between 0 and 5, [c]: There is a significant difference between 0 and 24, [d]: There is a significant difference between 1and 5, [e]: There is a significant difference between 1 and 24, [f]: There is a significant difference between 5 and 24

Tab. 4. Group II (mild pancreatitis) evaluation of serum biochemical values.

Variables	0th hour	1st hour	5th hour	24th hour	P value
Prolidase	755±10.02	773±11.7^{a}	778±12^{b}	785±16.6^{c}	< 0.001
Glucose	1.03±0.34	14.3±2.62^{a}	26±6.24^{bd}	50±5.46^{cef}	< 0.001
WBC	4.38±1.62	3.67±0.93	5.58±1.58^{d}	3.19±0.85^{f}	0.433
AST	93±16	143±27^{a}	188±34^{bd}	223±68^{c}	< 0.001
ALT	38±4.50	63±14^{a}	75±17^{b}	83±24^{c}	< 0.001
GGT	3.31±1.17	4.50±1.27	6.09±1.31^{b}	7.48±2.64^{ce}	< 0.001
ALP	57±12	149±20^{a}	196±34^{bd}	218±70^{c}	< 0.001
T. bil	0.12±0.03	0.13±0.05	0.18±0.04	0.11±0.31	0.596
D. bil	0.04±0.01	0.05±0.01	0.07±0.02^{b}	0.04±0.01^{f}	0.087
Amylase	70±20	1297±84^{a}	1691±103^{bd}	2293±309^{cef}	< 0.001
Lipase	7.66±2.16	11±1.18^{a}	15±2.91^{bd}	23±3.81^{cef}	< 0.001

[a]: There is a significant difference between 0 and 1, [b]: There is a significant difference between 0 and 5, [c]: There is a significant difference between 0 and 24, [d]: There is a significant difference between 1and 5, [e]: There is a significant difference between 1 and 24, [f]: There is a significant difference between 5 and 24

Tab. 5. Group III (severe pancreatitis) evaluation of serum biochemical values.

Variables	0th hour	1st hour	5th hour	24th hour	P value
Prolidase	764±11.7	776±10.9	784±10.5^{b}	792±10.6^{c}	< 0.001
Glucose	42±1.34	37±8.15	34±6.25^{b}	39±6.03	0.084
WBC	6.33±1.74	10±3.05	7.02±2.20	2.01±0.52^{cef}	< 0.001
AST	103±34	233±34	254±28^{b}	270±36^{c}	< 0.001
ALT	31.8±11	71±17^{a}	104±25^{bd}	122±25^{ce}	< 0.001
GGT	3.78±1.27	5.36±1.37	9.25±1.14^{bd}	8.79±1.90^{ce}	< 0.001
ALP	55±11	160±38^{a}	192±51b	222±36^{ce}	< 0.001
T. bil	0.58±0.13	0.11±0.02^{a}	0.10±0.03^{b}	0.09±0.03^{c}	< 0.001
D. bil	0.22±0.07	0.05±0.01^{a}	0.05±0.02^{b}	0.03±0.01^{cf}	< 0.001
Amylase	78±13	1642±431^{a}	5002±1458^{bd}	10000±0.01^{cef}	< 0.001
Lipase	14±4.66	32±9.33^{a}	451±45^{bd}	693±182^{cef}	< 0.001

[a]: There is a significant difference between 0 and 1, [b]: There is a significant difference between 0 and 5, [c]: There is a significant difference between 0 and 24, [d]: There is a significant difference between 1and 5, [e]: There is a significant difference between 1 and 24, [f]: There is a significant difference between 5 and 24

itself. In Group I, changes in serum prolidase levels were not statistically significant at beginning, 1st, 5th and 24th hour (Tab. 3) ($p > 0.05$). In Group II (Tab. 4) and Group III (Tab. 5) prolidase levels at beginning, 1st, 5th and 24th hour, showed a statistically significant increase when compared in each group itself ($p < 0.05$).

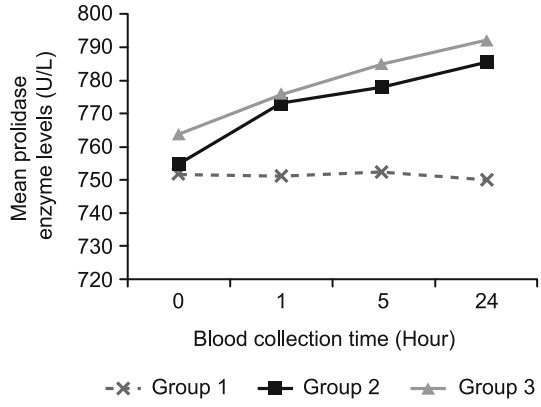

Fig. 2. Prolidase levels in groups at hours 0, 1, 5 and 24.

When we analysed the results between groups, serum prolidase levels in group II and group III were higher when compared with control group and this was statistically significant ($p < 0.05$) (Fig. 2). Comparison of prolidase levels in Group II with serum prolidase levels at beginning, 1st, 5th, and 24th hour in Group III showed an increase in general but there was no statistical significance ($p > 0.005$). In comparisons performed with tissue values, levels of prolidase in group II and group III were significantly higher

Tab. 6. Comparison of pancreatic tissue samples by groups.

Variables	Group I	Group II	Group III	p value
Prolidase	1665±188	1971±186^{a}	2217±245^{bc}	0.002

[a]: There is a significant difference between groups 1 and 2, [b]: There is a significant difference between groups 1 and 3, [c]: There is a significant difference between groups 2 and 3.

Tab. 7. Group II (50 µg cerulean) histopathological evaluation of rats.

GROUP	Edema	Inflammation	Necrosis
R-2-1	0	0	0
R-2-2	1	1	0
R-2-3	1	1	0
R-2-4	2	2	1
R-2-5	1	1	0
R-2-6	1	0	0
R-2-7	1	1	0
R-2-8	1	1	0
R-2-9	1	1	0
R-2-10	2	1	0

Tab. 8. Group III (80 µg cerulean) histopathological evaluation of rats.

GROUP	Edema	Inflammation	Necrosis
R-3-1	2	1	0
R-3-2	2	1	0
R-3-3	2	2	0
R-3-4	2	2	1
R-3-5	2	2	0
R-3-6	2	1	0
R-3-7	2	1	0
R-3-8	2	3	0
R-3-9	2	1	0
R-3-10	2	2	0

compared with the control group and this result was statistically significant ($p < 0.05$) (Tab. 6).

Histopathological evaluation

When groups examined histopathologically in our study, normal pancreatic tissue observed in group 1 (control group). One of the materials recorded in group 2 was normal and one had moderate and all others had mild pancreatitis. All of the samples recorded as group 3 had inflammation and edema but in two cases moderate pancreatitis was observed.

Microscopic examination of the groups revealed that, group 1 had normal pancreatic tissue. In group 2 acute inflammation and edema consisting of neutrophil polymorph and lymphocytes around ductus were observed (Tab. 7). In group 3, inflammation in the pancreatic tissue and focal area of acinar cell necrosis observed (Tab. 8).

Discussion

In 300 B.C. Herophilus identified the pancreas first time in history and 400 years later, "pancreas" described by Rufus. The definition of acute and chronic pancreatitis was made by Pare in 1579. In 1901, Opie has revealed the relationship between gallstones and pancreatitis and introduced anatomy of ampulla, choledoch and Wirsung and stated that their obstruction causes pancreatitis (9).

Many theories have been suggested about the pathophysiology of acute pancreatitis. In their study Beger and friends reported that in early and late phases of acute pancreatitis, deterioration of cell membrane and function occurs by the direct effect of reactive O_2 radicals and pancreas cell damage occurs by deregulation of lysosomal enzymes (10). Alhan et al found that after extravasation of pancreatic secretions into the interstitial space, proteolytic enzymes activated and initiated self-digestion process of tissue and as a result tissue edema, deterioration of microcirculation and cell ischemia develops. Lerch et al showed acinar cell necrosis within 3 hours, fat necrosis, hemorrhage and inflammation within 12 hours after the pancreatic duct ligation (11).

Extensive tissue distribution of collagen suggests that changes in prolidase enzyme activity which is a specific enzyme for its turnover, may be important for development of many diseases. In a few studies that evaluate prolidase enzyme activity in diseases that progress with chronic inflammation, prolidase enzyme activity was significantly higher due to collagen destruction (12,13,14).

Prolidase plays an important role in the transformation of prolin which involves recovery of imino acids from the proteins and stored collagen and has a role in collagen synthesis and cell growth (15). It is the only enzyme that functions in the destruction of dipeptides in the C-terminal of proline and hydroxyproline (X-proline or X-hydroxyproline) which occur in the last step of protein catabolism (16,17) and it is effective in the structure of the connective tissue and about 25 % of the continuity of the connective tissue (18).

Prolidase enzyme is responsible for breakdown of iminodipeptides that occur after collagen destruction. Increases in the collagen production and destruction process in fibrosis stage also causes increase in prolidase enzyme activity. Studies show that there is a correlation between serum prolidase levels and fibrotic activity.

Although in the literature there is a range of studies about prolidase enzymatic activity, there is no study about evaluation of prolidase activity which has a role in collagen metabolism in experimentally induced mild and severe pancreatitis in rats. In panreatic diseases that may affect many organ and tissue systems, it can be expected that prolidase activity, which also has an important role in production-destruction and reproduction of protein collagen, could be affected.

In our study, we used the Chinard method for measuring prolidase enzyme activity, which was modified first by Myara et al (19) and Özcan et al (20) afterwards (Chinard Method) (21).

In pancreatitis tissue edema, liquefaction, necrosis and bleeding occurs and in this process, activity of many enzymes such as elastase, kallikrein, phospholipase A, amylase and lipase, increases. Amylase secreted in its active form and break down carbohydrates. Lipase is activated by bile acids and break down lipids and leads to necrosis and in diagnosis, increase of this enzymes are used. Inexpensiveness, fastness, simplicity and avaibility of the test in so many places make amylase valuable. There are some studies suggesting that lipase is more sensitive and specific than amylase in diagnosis of acute pancreatitis (22, 23).

In their studies about acute necrotizing pancreatitis after i.p. injection of L-arginine on 32 rats, Czako et al (24) reported that serum amylase activity starts to increase and reached the highest level in the 24th hour and save a gradual decline in the subsequent hours, and reach the same values as the control group at the 48th hour. In our study also we showed a significant increase in amylase and lipase levels at beginning, 1st, 5th and 24th hour in group II (mild pancreatitis) and group III (severe pancreatitis) compared with the control group.

Çelik et al (25) have found the prolidase activity significantly lower in cirrhotic patients compared to the control group and suggested that collagen turnover has changed with the development of cirrhosis in human liver and prolidase activity may reflect disorders of collagen metabolism in this degenerative liver disease. Myara et al (26) reported in their studies that serum prolidase activity increases in liver cirrhosis and failure.

Alparslan et al (27) compared serum prolidase activity in three patient groups who have viral hepatitis, chronic hepatitis and cirrhosis with the control group in their study. These results suggest that serum prolidase activity can be a useful and independent variable in the diagnosis of liver disease and follow-up of chronic process.

Aslan et al (28) found that serum prolidase activity is significantly higher in H. pylori (+) patients compared with H. pylori (–) ones. They associated the prolidase activity with increase of collagen synthesis in cells and gastric fibrosis by gastric mucosal inflammation that was induced by oxidative stress in H. pylori (+) cases.

In their study, Altindag et al (29) found that serum prolidase activity was lower compared to the control group in patients with osteoarthritis of the knee and oxidative stress was increased. They interpreted this results as collagen metabolism may be decreased associated with oxidative stress.

Considering the studies indicating that inflammation creates changes in prolidase enzyme activity (28, 29, 30), one of the reasons of prolidase increase in patients with pancreatitis can be this increased

inflammatory state. As we know, reasons that held responsible for etiopathogenesis of pancreatitis (gallstones, alcohol and trauma, drugs, infections, metabolic reasons and free oxygen radicals); cause severe acinar cell injury, intense interstitial edema and hemorrhage in pancreatic tissue and as a result tissue inflammation occurs. In our study, a statistically significant increase of serum and tissue prolidase levels in group II and group III at beginning, 1st, 5th and 24th hour when compared to the control group, seem to support this data.

Neutrophilic and lymphocytic infiltration occurs histopathologically, in the development of acute pancreatitis. This inflammation of the gland tissue may lead to tissue destruction and even fibrosis later on. Meanwhile changes in collagen turnover, increase in prolidase enzyme activity, statistically significant increase compared to control group, all seem to support this data.

In conclusion, we want to clarify the relationship between acute pancreatitis and prolidase, to determine the affect of prolidase which is an important enzyme associated with collagen tissue, and also to shed light on future studies because we have not come across to a study on prolidase activity measurements in pancreatitis.

Because prolidase enzyme activity does not show large variations in healthy adults, this suggest that prolidase enzyme might be used for evaluation of collagen tissue damage in cases with acute pancreatitis. However, because lack of automated methods, it is not used as a routine parameter. Blood prolidase activity seems to be reliable parameter that will allow early diagnosis and follow-up in patients with collagen tissue damage if it is routinely used by full automatization of the method. Our study suggests that prolidase enzyme activity can be used as a feasible test for determine and control the damage process earlier in acute pancreatitis.

The findings obtained in our study showed that prolidase activity increases directly proportionaly with the severity of pancreatitis. This allows us the understanding that prolidase enzyme activities provide guidance about the metabolism of collagen in patients with acute pancreatitis, serious damage occurs in collagen protein and metabolic control is further distorted depending on the duration and intensity of damage but to be able to speak more precisely, there is a need for further, more detailed and extensive researchs.

References

1. Thomson SR, Hendry WS, McFarlane GA, Davidson AI. Epidemiology and Outcome of Acute Pancreatitis. Br J Surg 1987; 74: 398–401.

2. Steer ML, Meldolesi J, Figarella C. Pankreatitis. The role of lysosomes. Dig Dis Sci 1984; 29: 934–938.

3. Myara I. Effect of long preincubation on the two forms of human erythrocyte prolidase. Clin Chim Acta 1987; 170 (2–3): 263–270.

4. Milligan A, Brown G. Prolidase Deficiency: A Case Report and Literature Review. Brit J Dermatol 1989; 121: 405–409.

5. Endo F, Matsuda I. Human eritrosite Prolidase and prolidase deficieny. Pediatr Res 1982; 16: 227–231.

6. Mayer J, Rau B, Schoenberg MH et al. Mechanism and role of trypsinogen activation in acute pancreatitis. Hepato-Gastroenterol 1999; 46: 2757–2763.

7. Schoenberg MH, Buchler M, Beger HG. Oxygen radicals in experimental acute pancreatitis. Hepatogastroenterol 1994; 41: 313–319.

8. Schmidt J, Rattner DW, Lewandrowski K, Compton CC, Mandavilli U, Knoefel WT, and Warshaw AL. A better model of acute pancreatitis for evaluating therapy. Ann Surg 1992; 215: 44–56.

9. Banks PA. Epidemiology, natural history, and predictors of disease outcome in acute and chronic pancreatitis. Gastrointest Endosc 2002; 56 (Suppl): S226–230.

10. Beger HG, Rau BM. Severe acute pancreatitis: Clinical course and management. World J Gastroenterol 2007; 13: 5043–5051.

11. Lerch MM, Saluja AK, Dawra R, Ramaraò P, Saluja M, Steer ML. Acute necrotizing pancreatitis in the opossum: earliest morphological changes involve acinar cells. Gastroenterology 1992; 103: 205–213.

12. Oono T, Fujiwara Y, Yoshioka T, Arata J. Prolidase activity in chronic wound and blister fluids. J Dermatol 1997; 24 (10): 626–635.

13. Aslan M, Nazligül Y, Horoz M, Bölükbaş C, Bölükbaş F, Aksoy N, Çelik H, Erel Ö. Serum prolidase activity and oxidative status in helicobacter pylori infection. Clin Biochem 2007; 40: 37–40.

14. Soner Y, Gurdol F, Tuğrul Y, Bekpinar S. Prolidase I activity in liver tissue: effects of ethanol and selenium. Res Commun Alcohol Subs Abuse 1995; 16: 125.

15. Atara J, Umemura S, Yamamato Y, Hagiyama M, Nohara N. Prolidase deficiency: Its dermatalogical manifestations and some additional biochemical studies. Arch Dermatol 1979; 115: 62.

16. Endo FA, Matsuda I. Molecular basis of prolidase (peptidase D) deficiency. Mol Biol Med 1991; 8: 117–127.

17. Myara I, Cesson C, Moatti N, Lemonnier A. Human kidney prolidase-purification, preincubation properties and immunological reactivity. Int J Biochem 1994; 26: 207–214.

18. Kurien BT, Patel NC, Porter AC, D'Souza A, Miller D, Matsu-moto H, Wang H, Scofield RH. Prolidase deficiency and the biochemical assays used in its diagnosis. Anal Biochem 2005; 349 (2): 165–175.

19. Myara I, Charpentier C, Lemonnier A. Optimal conditions for prolidase assay by proline colorimetric determination: Application to iminodipeptiduria. Clin Chim Acta 1982; 125: 193–205.

20. Özcan Ö, Gültepe M, İpçioğlu OM, Bolat B, Kayadibi H. Optimization of the Photometric Enzyme Activity Assay For Evaluating Real Activity of Prolidase]. Turk J Biochem 2007; 32 (1): 12–16.

21. Clavien PA, Burgan S, Moossa AR. Serum enzymes and other laboratory tests in acute pancreatitis. Br J Surg 1988; 76: 1234–1243

22. Eckfeldt JH, Levitt MD. Diagnostic enzymes for pancreatic disease. Clin Lab Med 1989; 9: 731–734.

23. Gumaste VV, Roditis N, Metha D, Dave PB. Serum lipase levels in non-pancreatic abdominal pain versus acute pancreatitis. Am J Gastroenterol 1993; 88: 2051–2054.

24. Czako L, Takacs T, Varga IS et al. The pathogenesis of L-arginine induced acute necrotizing pancreatitits: Inflammatory mediators and endogenous cholecystokinin. J Physiol Paris 2000; 94: 43–50.

25. Çelik H, Aksoy N, Aslan M, Naligül Y, Barut Ş. Disregulation of Collagen Metabolism in Cirrhotic Patients. Turk J Biochem 2005; 29 (1): 1–172.

26. Myara I, Myara A. Plasma prolidase activity: A possible index of collogen catabolism in chronic liver disease. Clin Chem 1984; 30 (2): 211–215.

27. Alparslan S, Gültepe M. Serum prolidase activity: its value as an indicator of collagen accumulation in chronic liver diseases. Biochem J 1993; 18: 1–9.

28. Aslan M, Nazligül Y, Horoz M, Bölükbaş C, Bölükbaş F, Aksoy N, Çelik H, Erel Ö. Serum prolidase activity and oxidative status in helicobacter pylori infection. Clin Biochem 2007; 40: 37–40.

29. Altindag O, Erel O, Aksoy N, Selek S, Celik H, Karaoglanoglu M. Increased oxidative stress and its relation with collagen metabolism in knee osteoarthritis. Rheumatol Int 2007; 27 (4): 339–344.

30. Kaleli S, Akaya A, Akdoğan M, Gültekin F. The Effects of different treatments on prolidase and antioxidant enzyme activities in patients with bronchial asthma. Environ Toxicol Pharmacol 2006; 22: 35–39.

Received July 19, 2016.
Accepted August 4, 2016.