



A Comparative Study of Rapid Trypsinogen 2 Strip Test Versus Serum Amylase and Serum Lipase in Diagnosis of Acute Pancreatitis in A Tertiary Care Hospital, Salem

Arun Balaji¹, Subalakshmi.P^{2*}, A.P. Subburaj³, J. Sridhar⁴, E.M.J Karthikeyan⁵,
Kumaran.K¹

Assistant Professor¹, Postgraduate², Professor³, Professor and HOD⁴, Department of General Surgery, Director⁵, Vinayaka Mission's Kirupananda Variyar Medical College & Hospital, Vinayaka Mission's Research Foundation (DU), Salem (Tamil Nadu), India.

*Corresponding author's E-mail: mbsubalakshmi@gmail.com

Article History	Abstract
Received: 06 June 2023 Revised: 05 Sept 2023 Accepted: 11 Sept 2023	<p>Aims & Objectives: To assess the diagnostic usefulness of a quick urine trypsinogen 2 strip test to blood amylase and lipase in instances with acute pancreatitis. Methods: Patients who may have an acute pancreatitis diagnosis and have recent onset of stomach pain are included. In these cases, serum samples of amylase and lipase are sent before a urine sample is examined using a dipstick test for urinary trypsinogen-2. Fifty-two participants were included in the research. Ultrasound and CECT abdomen were used to monitor patients and confirm the diagnosis. A number of metrics were computed and compared, including sensitivity, specificity, PPV, NPV, and accuracy. Result: During June 2022 and June 2023, 52 consecutive patients complaining of upper abdominal discomfort were included in the research. Acute pancreatitis was the diagnosed diagnosis in 45 of the 52 patients, whereas other causes of abdominal discomfort were present in 7 individuals. The pee dipstick test was positive in 44 of the 45 instances, but was negative in 5 of the 7 cases when the discomfort in the abdomen was caused by anything other than a urinary tract infection. It was shown to have a 97.78 percent sensitivity, a 71.43 percent specificity, and a 94.23 % accuracy. Conclusion: Acute pancreatitis may be diagnosed quickly and easily with a fast urine trypsinogen-2 test. This study shows that the dipstick test is just as accurate as the gold standard procedures and may be used alternately with them.</p>
CC License CC-BY-NC-SA 4.0	Keywords: Trypsinogen, Serum Amylase, Serum Lipase, Pancreatitis, Dipstick test

1. Introduction

The most common reason for severe stomach pain that sends patients to the emergency department is acute pancreatitis. Acute pancreatitis is a condition characterized by an inflammatory response centered on the pancreas but often spreading to the surrounding tissues and even other organs (Brunnicardi, 2018). While the majority of patients suffer a mild, self-limited course that resolves on its own, 10 percent to 20 percent of patients develop a rapidly escalating inflammatory response that is linked with a longer hospital stay, significant morbidity, and even mortality (Kemppainen, 1997 & Corfield, 1985). Acute pancreatitis must be diagnosed quickly so that it may be treated effectively and quickly.

Misdiagnosis of acute pancreatitis is possible due to its shared clinical characteristics with other acute abdominal diseases. The detection of amylase in serum or urine is one of the key laboratory tests for identifying acute pancreatitis, yet hyperamylasemia is absent in 19% of patients (Clavien, 1989).

Patients with acute abdominal discomfort that is not caused by the pancreas may see an elevation in these pancreatic enzyme levels.

Trypsinogen levels in the blood are a good indicator of pancreatic damage, and proteolytic enzymes are very important in the development of AP. Three isoforms of trypsinogen, a pancreatic proteinase with a 25-kilodalton (kd) molecular weight, are released by the human pancreas. The two main isoenzymes, trypsinogen-1 and trypsinogen-2, are present in considerable quantities in pancreatic fluid, but very little of this fluid enters the circulation (Itkonen, 1990). Trypsinogens are easily filtered by the glomeruli because of their tiny size. Trypsinogen-2 has a larger urine concentration than trypsinogen-1 because, for reasons that have yet to be determined, its tubular re absorption is lower.

A crucial step in the pathophysiology of acute pancreatitis is the conversion of trypsinogen to trypsin inside the pancreas (Marshall, 1993). We present the findings of a prospective research that evaluated the dipstick's diagnostic value for acute pancreatitis.

Aims And Objectives:

Aim: In cases of acute pancreatitis, compare the diagnostic value of a rapid urine trypsinogen 2 strip test to that of blood amylase and lipase.

Objectives: We assessed the sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) of urine trypsinogen to blood amylase and lipase as well as the gold standard of CT scan (NPV).

2. Materials And Methods

A prospective analytical study was conducted between June 22 and June 23, 2012, at the Kirupananda Variyar Medical College and Hospital of the Vinayaka Mission in Salem. The study comprised patients who were hospitalised to our hospital with severe upper abdominal pain. The calculated sample size was 42 patients. Including the drop outs we included 52 patients in our study.

Criteria:

Patients over the age of 18 who presented with severe upper abdominal pain and gave their verbal and written consent to participate were generally considered acceptable. Patients who met the exclusion criteria were those who were less than 18 years old or who had ERCP-induced pancreatitis.

Technique and Protocol:

All patients who came to the Department of General Surgery and satisfied the inclusion criteria were enrolled for this prospective analytical research. A complete history and physical examination were performed on each patient from the outset. Acute pancreatitis was suspected since the patient had just started drinking, had a history of anorexia and vomiting, and was experiencing significant stomach pain that was radiating to the back.

Specimen Collection: At the time of admission, UTDT was used to measure trypsinogen-2 in a urine sample.

Trypsinogen-2 Measurements: An immunochromatographic test known as a dipstick test may be used to assess urinary trypsinogen2. Trypsinogen-2-bound blue latex particles migrate across a nitrocellulose membrane after the test strip has been dipped into the urine sample and has been labelled with a second antibody that is specific for a separate trypsinogen-2 epitope. A blue line appears in this area when the concentration of trypsinogen-2 is more than 50 ng/mL The urine-filled vial was held over the strip's tip for 20 seconds before the strip was entirely removed. After that, the strip sat out for 5 minutes at room temperature.

For the research, sonographic findings were reported as positive (indicating the presence of acute pancreatitis) or negative (indicating the absence of this condition). Patients were observed, and a CECT of the abdomen and pelvis was carried out to confirm the diagnosis and determine the severity of the problem when pancreatitis was suspected. The CECT result was either regarded as positive or negative for pancreatitis since this study is a qualitative test for acute pancreatitis screening.

A blood lipase content that is more than three times the typical upper limit (180 U/L) is indicative of hyperlipaemia. The acute pancreatitis symptom panel includes two of the three symptoms listed below:

1. Acute pancreatitis symptoms include severe abdominal pain.
2. Extremely high levels of amylase and/or lipase in the plasma
3. characteristic CT scan results

3. Results and Discussion

A prospective, analytical study on patients undergoing general surgery was conducted from June 2022 to June 2023 at the Kirupananda Variyar Medical College and Hospital of the Vinayaka Mission in Salem, India. Patients who sought medical attention for sudden onset of upper abdomen discomfort throughout the research period were included.

The research involved 52 consecutive patients with upper abdomen discomfort who met both inclusion and exclusion criteria. The youngest patient with acute pancreatitis was 18 years old, and in the non-pancreatic cause of abdominal pain group, the youngest was 23 years old. On average, patients with acute pancreatitis were 84 years old, while those with other causes of abdominal discomfort were, on average, 59 years old. figure 1 shows the age distribution in acute pancreatitis group and non- pancreatic cause of pain abdomen.

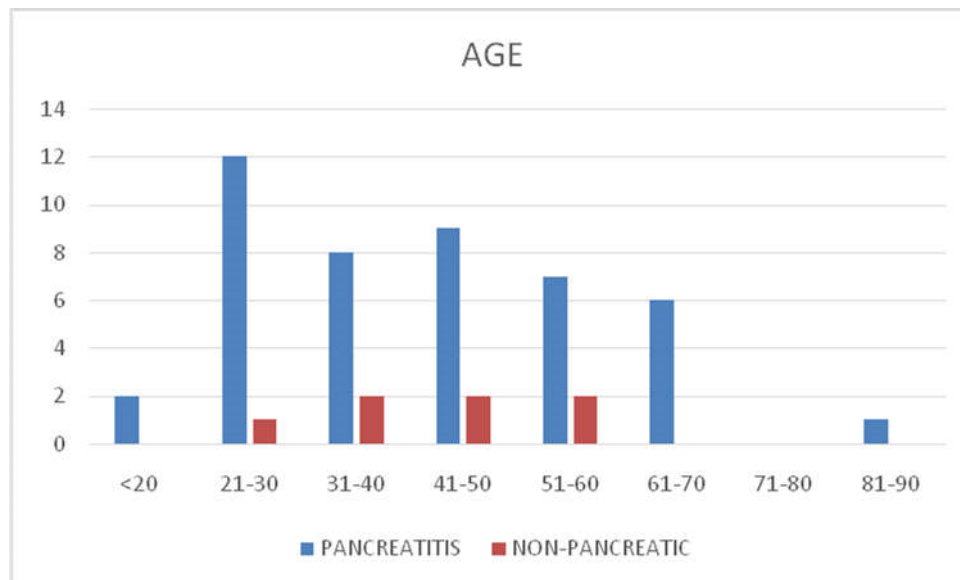


Figure 1: Age Distribution in each group

Males were more likely than women to have upper abdominal discomfort, with 44 men reporting it vs 8 women (84.6 percent vs. 15.4 percent, respectively (Figure 2). The gender distribution of patients with acute pancreatitis is 39 men and 6 females, while the gender distribution of patients with non-pancreatic stomach discomfort is 5 males and 2 females. This suggests that acute pancreatitis is more common in men. Study results shows that among the patients who had stomach discomfort that was not caused by pancreatitis, 42.8% had acute gastritis, 42.8% had acute calculous cholecystitis, and 14.8% had choledocholithiasis.

Pancreatitis is the most common cause of stomach discomfort in patients, and our analysis of these cases revealed the following distribution of causes. Of the 45 instances of pancreatitis identified in our investigation, alcohol was a major risk factor in 29 of them. According to table 9 and figure 11, gallstone disease was the second leading cause, accounting for 24.4%. This was followed by idiopathic causes, which accounted for 6.6%, and elevated TGL, which accounted for 4.4% (Table-1).

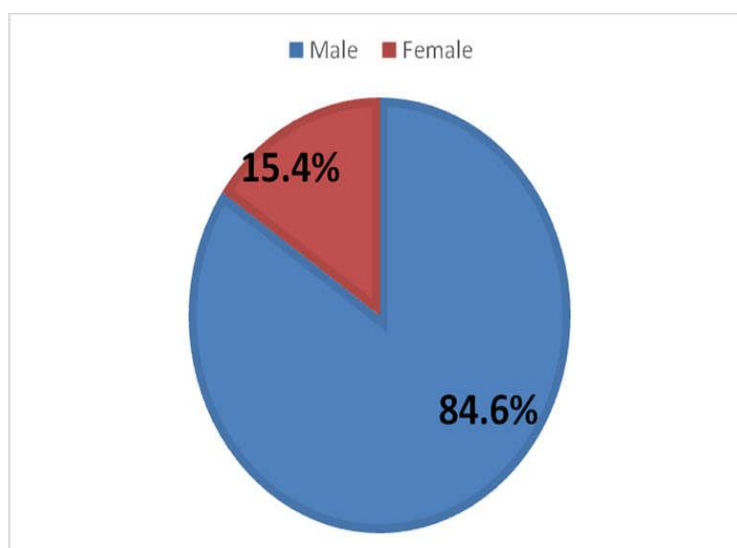


Figure 2: Gender Distribution

Table 1: Aetiology of Acute Pancreatitis

Aetiology	Frequency	Percentage
Alcohol	29	64.4%
Gallstone	11	24.4%
Idiopathic	3	6.6%
Hypertriglyceridemia	2	4.4%

We intended to evaluate the sensitivity, specificity, accuracy, positive predictive value, and negative predictive value of the various investigations once data collection was completed.

Table 2 showed that S. Amylase had an 88.4% accuracy, a 95.3 percent positive predictive value, a 55.5 percent negative predictive value, and a sensitivity of 91.1 percent. Table 10a shows that S. Amylase has a 91.1 % sensitivity, a 71.4 % specificity, a 95.3 % positive predictive value, a 55.5 % negative predictive value, and an 88.4 % accuracy.

Table 2: Analysis of Serum Amylase

SERUM AMYLASE	ACUTE PANCREATITIS	NON -PANCREATIC
POSITIVE	41	2
NEGATIVE	4	5

Table-3 shows the serum lipase has an 85.7 percent specific and a 93.3 percent sensitivity for accurate diagnosis. Table 10b shows that it had a positive predictive value of 97.7 percent, a negative predictive value of 66.6, and an accuracy of 92.3 percent.

Table 3: Analysis of Serum Lipase

SERUM LIPASE	ACUTE PANCREATITIS	NON -PANCREATIC
POSITIVE	42	1
NEGATIVE	3	6

USG was shown to have an 85.7 percent specificity rate and a 53.3 percent sensitivity rate. It was 57.6 percent accurate, had a 96 percent positive predictive value, and a 22.6 percent negative predictive value. We believe that it has certain impediments because the pancreas may not be visualized due to bowel gas and extra pancreatic spread of inflammation and vascular inconveniences may not be picked up by Ultrasonography. Our analysis of the data showed that CECT Abdomen has a perfect sensitivity, specificity, PPV, and NPV, as well as an accuracy rate of 100%. (Table-4)

Table 4: Representation of all parameters

Test	Sensitivity	Specificity	PPV	NPV	Accuracy
Amylase \geq 3 ULN	91.1	71.4	95.3	55.5	88.4
Lipase \geq 3ULN	93.3	85.7	97.7	66.6	92.3
USG	53.3	85.7	96	22.2	57.6
CECT	100	100	100	100	100
UTDT	97.7	71.4	95.6	83.3	94.2

Most cases of acute pancreatitis are brought on by either bile stone formation or heavy alcohol usage. Fluid resuscitation, pain control, and early initiation of oral feeds usually result in a quick clinical recovery since the clinical course is often moderate. There is a severe variant of the illness that affects 20-30% of people and has a death rate of about 15% (Elman, 1929). Around half of all occurrences of acute pancreatitis in the UK are still caused by gallstone disease, which is still the most frequent cause (Shah, 2018). Gallstone disease, hypertriglyceridemia, and idiopathic were the other possible explanations. In the non-pancreatic causes of pain abdomen group, the two most common causes were acute acalculous cholecystitis and acute gastritis.

Since they are affordable and offer a good mix of sensitivity and specificity, pancreatic enzyme tests such blood amylase and lipase are the most often used indications for early identification of acute pancreatitis (Treacy, 2001). At this time, there is no gold standard marker for the treatment of acute pancreatitis that enables precise diagnosis, early prediction of the course of the disease, and cause identification. Although blood amylase and lipase levels may rise as a result of intra-abdominal inflammation and hypertriglyceridemia, this has the effect of lowering the sensitivity and specificity of the tests used to identify acute pancreatitis. A similar limitation is seen in renal insufficiency as 25% of amylase is cleared off by the kidneys (Smotkin, 2002 & Kylänpää-Bäck, 2000).

The present investigation found that serum amylase and serum lipase had respective values of 91.1%, 71.4%, 95.3%, 55.5%, and 93.3%, 85.7%, 97.6%, and 66.6% for sensitivity, specificity, PPV, and NPV. The findings showed that serum lipase had a greater accuracy of 92.3 percent compared to 88.4 percent when the diagnostic accuracies of the enzymatic tests were assessed. Although the glomeruli can easily filter both of the trypsinogen isoenzymes, trypsinogen-1 and trypsinogen-2, trypsinogen-2 is selectively enhanced in acute pancreatitis. Trypsin activation inside the pancreas is a more severe version of the illness that characterises late-stage AP. A simple urine dipstick test for trypsinogen-2 has been proposed as a potential early diagnostic tool for acute pancreatitis. To measure the concentration of trypsinogen-2 in the urine, we employed a dipstick test (Action Pancreatitis, Medix Biochemica Oy AB, Kauniainen, Finland) based on an immunochromatography assay. The dipstick method was a simple test to carry out and results were attained within 5 minutes.

Previous research has shown that UTDT's sensitivity is between 53.3% and 96.0%, and its specificity is between 85.7% and 95.0% (Kamer, 2007). We determined that UTDT has a 97.7 percent sensitivity and a 71.4 percent specificity. Consistent with prior research, we discovered that UTDT had a better sensitivity than serum amylase and serum lipase (97% vs. 91.1% and 93.3%, respectively) (Lempinen,2001). 59 The steady rise in urine trypsinogen 2 levels in acute pancreatitis may account for the test strip's excellent clinical sensitivity (Munoz, 2000).

This study finds that the PPV (95.6%) for UTDT was somewhat lower than that of serum lipase but almost identical to that of serum amylase. A urine screening test may be beneficial for patients who visit the emergency department since it may lessen the possibility that acute pancreatitis may go undiagnosed. Our results suggest that the UTDT strip should be given this function. Acute pancreatitis may be ruled out with high confidence in the absence of positive findings, while positive results often point to patients who need more testing. Abdominal imaging is helpful for confirming the diagnosis of AP. When compared to CT, ultrasound of the abdomen has a lower sensitivity and specificity for identifying necrosis inside the pancreas and outlining peripancreatic inflammation". When it comes to making a diagnosis of AP, CECT has a sensitivity and specificity of about 90%. Our research found a

100% sensitivity and specificity. Since most people have a mild, uneventful course, CECT is not necessary for everyone.

CECT is indicated to determine the severity of the condition if the patient does not show signs of improvement within 48 to 72 hours.

Finally, the UTDT's findings were straightforward to interpret and had the benefit of instant feedback. Serum amylase and lipase tests, which are frequently performed, did not provide a perfect result, but they were nonetheless conclusive.

4. Conclusion

UTDT exhibited a somewhat higher sensitivity than serum amylase and serum lipase, the researchers concluded after analysing the data and doing statistical analysis, as was previously mentioned. Using urine Trypsinogen-2 dipsticks, the majority of cases of acute pancreatitis may be either identified or ruled out with a simple, rapid, easy, and non-invasive test. It is possible to estimate the trypsinogen-2 concentration in urine without using a lab. This test is completed fairly quickly in contrast to serum amylase and lipase, which may take an hour or more to get findings (within 5 minutes). It cannot be used as a diagnostic test for acute pancreatitis due to its low specificity, but it can be helpful as a screening test. The study had a small sample size, an observational study and there were fewer cases in the non-pancreatic cause of abdominal discomfort group need to do large group of study across the country.

Acknowledgments: We sincerely thank each and every one of the participants for taking part in research study. The authors thank the Vinayaka Mission's Kirupananda Variyar Medical College and Hospital, Salem, Tamil Nadu, for providing the necessary facilities.

Conflict of interest: Nil

Author's contribution: **Dr Arun Balaji M S** - conceptualization, data curation, investigation, methodology, project administration, visualization, writing—original draft, writing—review and editing; **Prof. Dr A.P. Subburaaj** -conceptualization, methodology, writing—original draft, writing—review and editing; **Prof. Dr. J. Sridhar** - conceptualization, visualization, supervision, writing—original draft; **Prof. Dr. E.M.J Karthikeyan** - project administration, original draft review and editing. **Dr Kumaran.K** and **Dr Subalakshmi.P** - methodology, writing—original draft, writing, review and editing. All authors approved the final manuscript as submitted and agree to be accountable for all aspects of the work. All authors have read and agreed to the published version of the manuscript.

Data Availability: All datasets generated or analysed during this study are included in the manuscript.

IEC Approval: Institutional Ethics Committee Approval from Vinayaka Mission's Kirupananda Variyar Medical College and Hospital, Salem, Tamil Nadu. Written informed consent was obtained from the participants before enrolling in the study.

References:

- Brunnicardi FC, editor. (2018). *Schwartz's principles of surgery* (Eleventh edition). New York: McGraw-Hill.
- Clavien PA, Robert J, Meyer P, Borst F, Hauser H, Herrmann F, et al. (1989). Acute pancreatitis and normoamylasemia. Not an uncommon combination. *Ann Surg*, 210(5), 614–620.
- Corfield AP, Williamson RCN, McMahon MJ, Shearer MG, Cooper MJ, Mayer AD, et al. (1985). Prediction of severity in acute pancreatitis: prospective comparison of three prognostic indices. *The Lancet*, 326(8452), 403–407.
- Elman R. (1929). Value Of Blood Amylase Estimation in the diagnosis of pancreatic disease: a clinical study. *Arch Surg*, 19(6), 943.
- Itkonen O, Koivunen E, Hurme M, Alfthan H, Schröder T, Stenman UH. (1990). Timeresolved immunofluorometric assays for trypsinogen-1 and 2 in serum reveal preferential elevation of trypsinogen-2 in pancreatitis. *The Journal of Laboratory and Clinical Medicine*, 115(6), 712–718.
- Kamer E, Unalp HR, Derici H, Tansug T, Onal MA. (2007). Early diagnosis and prediction of severity in acute pancreatitis using the urine trypsinogen-2 dipstick test: A prospective study. *World J Gastroenterol*, 13(46), 6208–6212.

- Kemppainen EA, Hedström JI, Puolakkainen PA, Sainio VS, Haapiainen RK, Perhoniemi V, et al. (1997). Rapid Measurement of Urinary Trypsinogen-2 as a Screening Test for Acute Pancreatitis. *N Engl J Med*, 336(25), 1788–1793.
- Kylänpää-Bäck M-L, Kemppainen E, Puolakkainen P, Hedström J, Haapiainen R, Perhoniemi V, et al. (2000). Reliable screening for acute pancreatitis with rapid urine trypsinogen-2 test strip: Screening for acute pancreatitis. *Br J Surg*, 87(1), 49–52.
- Lempinen M, Kylänpää-Bäck M-L, Stenman U-H, Puolakkainen P, Haapiainen R, Finne P, et al. (2001). Predicting the Severity of Acute Pancreatitis by Rapid Measurement of Trypsinogen-2 in Urine. *Clinical Chemistry*, 47(12), 2103–2107.
- Marshall JB. (1993). Acute Pancreatitis: A Review With an Emphasis on New Developments. *Arch Intern Med*, 153(10), 1185.
- Munoz A, Katerndahl DA. (2000). Diagnosis and Management of Acute Pancreatitis. *AFP*, 62(1), 164–174.
- Shah AP, Mourad MM, Bramhall SR. (2018). Acute pancreatitis: current perspectives on diagnosis and management. *J Inflamm Res*, 11, 77–85.
- Smotkin J, Tenner S. (2002). Laboratory Diagnostic Tests in Acute Pancreatitis. *Journal of Clinical Gastroenterology*, 34(4), 459–462.
- Treacy J, Williams A, Bais R, Willson K, Worthley C, Reece J, et al. (2001). Evaluation of amylase and lipase in the diagnosis of acute pancreatitis: amylase and lipase in acute pancreatitis. *ANZ Journal of Surgery*, 71(10), 577–582.