

Research Article

The Correlation Study Between Seminal Fluid Enzymes and Effects on Men's Fertility

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Received: 15 April, 2023

Accepted: 19 May, 2024

Published: 25 May 2024

Abstract:

Objective: By analyzing the relationship between ALP, GOT, and LDH action within the seminal plasma of fruitless men and comparing it with ripe men, this study aimed to investigate the physiological conditions that remain unexplained in most cases.

Methods: This study collected forty-four semen tests from normospermic men and 20 tests from fertile men, after a period of forbearance (3-5 days). The tests were analyzed within the Babylon Maternity and Children research facility and private research facilities, from January 2024 to April 2024.

Results: The results indicated that ALP and AST activity had a negative correlation in both fertile ($r = -0.123$) and normospermic men ($r = -0.038$). Similarly, ALP and LDH activity had a negative correlation in fertile men ($r = -0.334$), but a positive correlation that was not significant ($P > 0.05$) in normospermic men ($r = 0.128$). Moreover, the results showed that AST and LDH activity positively correlated in both normospermic ($r = 0.058$) and fertile men ($r = 0.242$).

Conclusion: The study hypothesized that normal semen components and gland secretory ability may be indicated by ALP, AST, and LDH activity. The levels of ALP, AST, and LDH activity in males who were fertile and those who were not showed any discernible differences.

Keywords: Alkaline phosphatase (ALP), Aspartate aminotransferase (AST), lactate dehydrogenase (LDH) and normospermia.

1- Introduction

A crucial component of human semen, seminal plasma aids in the survival and migration of sperm in the female reproductive system (1). It contains a variety of components and enzymes, such as alkaline phosphatase (ALP, EC 3.1.3.1), aspartate transaminase (AST, EC 2.6.1.1), and lactate dehydrogenase (LDH, EC 1.1.1.27), among others. These enzymes are essential for the metabolic processes that provide energy for the survival, activity, and motility of spermatozoa (2).

The epididymis and testis are the primary sources of the high concentration of ALP seen in the seminal plasma. Four times as much ALP is present in seminal plasma as in human, bovine, and rabbit sperm (3). Aspartate aminotransferase is another enzyme that is abundant in the seminal plasma and may be found in many different human tissues and organs. This enzyme is mostly found in the seminal plasma of the prostate gland, and it plays a major role in the overall activity of the human ejaculate (4).

According to recently study for several metabolic processes that

provide energy for spermatozoa survival, motility, and fertility, the enzyme lactate dehydrogenase (LDH) is necessary. Inside the cells, nicotinamide-adenine dinucleotide (NAD)⁺ serves as a hydrogen acceptor. This enzyme transfers a hydrogen atom, which catalyzes the reverse process of turning L-lactate into pyruvate (5).

The primary source of LDH activity in seminal plasma is the prostate, and the sperm-specific isoenzyme LDH-C4 accounts for around 30% of this activity (6).

The existence or lack of sperm and the physiological state of men are correlated with the enzyme activity in the seminal plasma. Male infertility has been linked to the enzyme activity in seminal plasma in several studies (7). Congenital defects, and external influences including anabolic steroids, smoking, alcohol, pharmaceuticals, and other variables are some of the causes of male infertility. Approximately 40–50% of male infertility cases are caused by these variables (8).

Based on the characteristics of the semen following seminal fluid analysis, the World Health Organization (WHO) categorized the patients with infertility (9).

2-Material and method

2-1 Activity of alkaline phosphatase (ALP):

Alkaline phosphatase (ALP) activity in the seminal plasma was measured using the kinetic technique and a kit from Liner Chemicals, a Cromatest Spanish business.

2-2 Activity of Aspartate-Aminotransferase (AST):

Using a kit from Agappe (a Swiss business), the kinetic approach was utilized to evaluate the activity of aspartate-aminotransferase (AST) in the seminal plasma.

2-3 Activity of lactate dehydrogenase (LDH):

Lactate dehydrogenase (LDH) activity in the seminal plasma was measured by the kinetic technique using a kit from Bio Maghreb (a Tunisian business).

2-4. Getting specimens ready for LDH evaluation:

The sperm and round cells were separated from the seminal plasma by centrifuging the semen samples twice, for ten minutes each time, at 3000 rpm. Following the initial centrifugation, the supernatant, or seminal plasma, was transferred to a fresh tube and stored at -20 °C until the test.

2-5 samples of semen:

After refraining from sexual activity for three days, the patients gave the samples by having masturbation. After the specimens were liquefied at 37 °C, the sperm parameters were examined. Ten samples of fertile males were also included, and the infertile patients were categorized into normospermia (22 samples) based on the study.

2-6: Analysis of statistics:

The data was analyzed using the statistical software for Social Science (SPSS) version 16.0. To determine whether there were any significant differences between the groups, the mean \pm S.D. for each group was determined and the Duncan experiment design was used. Additionally, linear relationships between the activity of the enzymes were found (10).

3-Results

3-1 The degree of enzyme activity

Between normospermic patients and fertile males, (Fig 1) displays non-significant changes ($P > 0.05$) in the seminal plasma ALP activity.

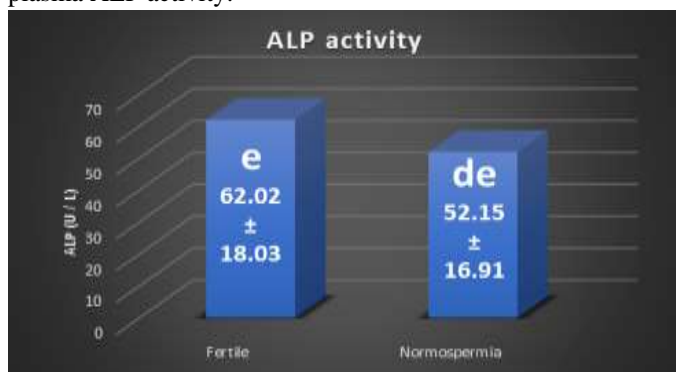


Figure-1 Alkaline phosphatase activity levels letters mean significant difference. (mean \pm S.D).

The seminal plasma of normothermic patients and fertile males was shown to have AST activity in (Fig 2), which did not significantly vary ($P > 0.05$) between the two groups.

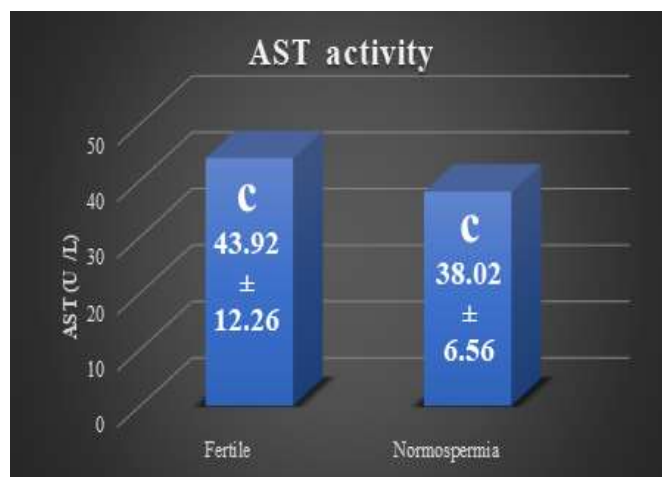


Figure-2 Aspartate aminotransferase activity levels

(Fig 3) displays the degree of LDH activity in the seminal plasma of males with normospermia and those who are fertile, noting non-significant differences ($P > 0.05$).

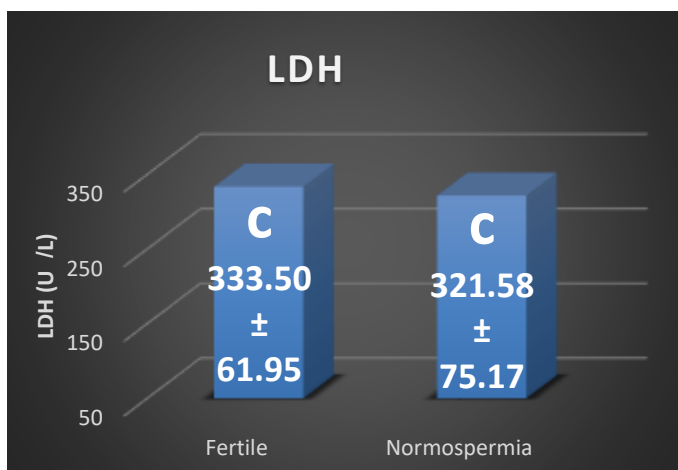


Figure-3 Lactate dehydrogenase activity levels.

3-2 The correlations of enzymes:

The findings showed that ALP and AST activity in the groups with normothermia ($r = -0.038$) and fertile individuals ($r = -0.123$) had negative relationships (Figs. 4 & 5).

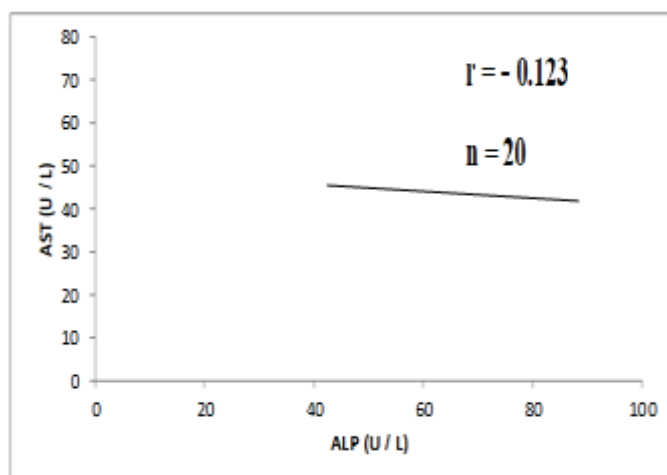


Figure 4 displays the relationship between AST and ALP activity in fertile individuals' seminal plasma.

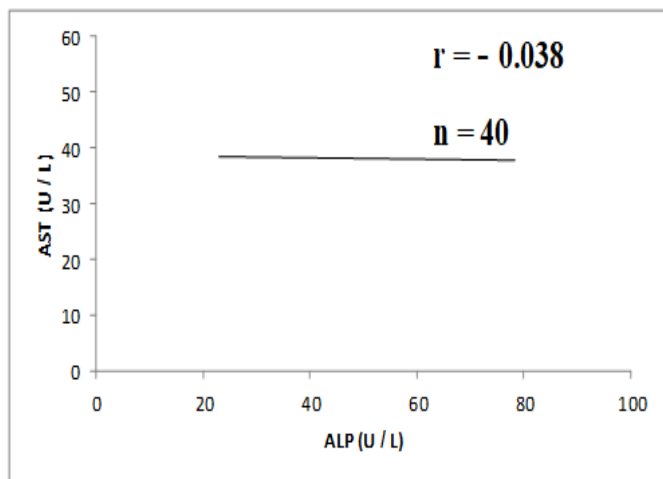


Figure 5 displays the relationship between ALP & AST activity in normospermic seminal plasma.

Furthermore, contrasted to negative correlations in fertile men ($r = -0.335$) (figure 7), the data demonstrated a positive connection between ALP and LDH activity in the seminal plasma of normothermic patients ($r = 0.128$) (Fig. 6).

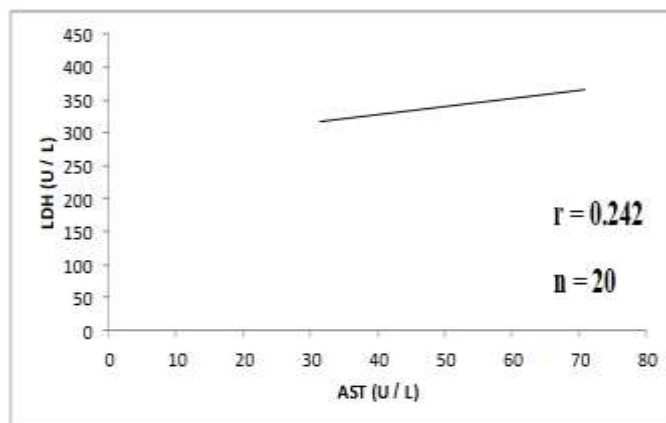


Figure 8 The relationship between AST &LDH activity in the seminal plasma of fertile individuals

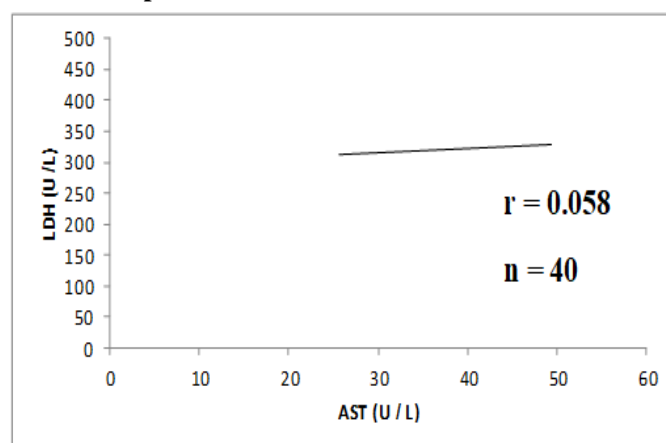


Figure 9 The correlation between the seminal plasma of normospermia &LDH activity

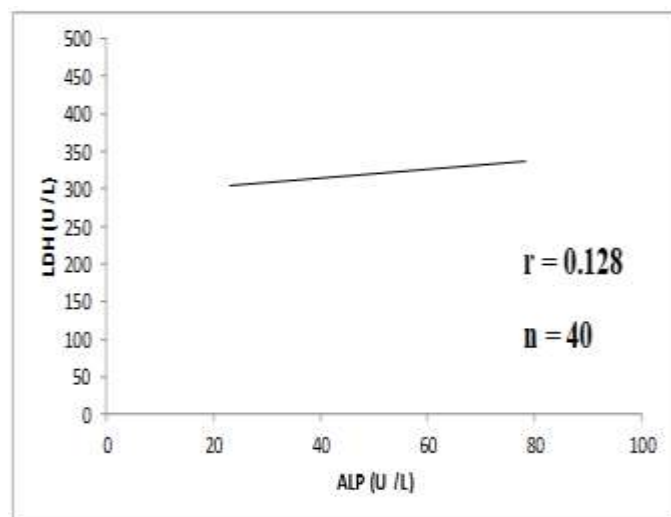


Figure 6 The relationship between ALP and LDH activity in the seminal plasma of normospermia

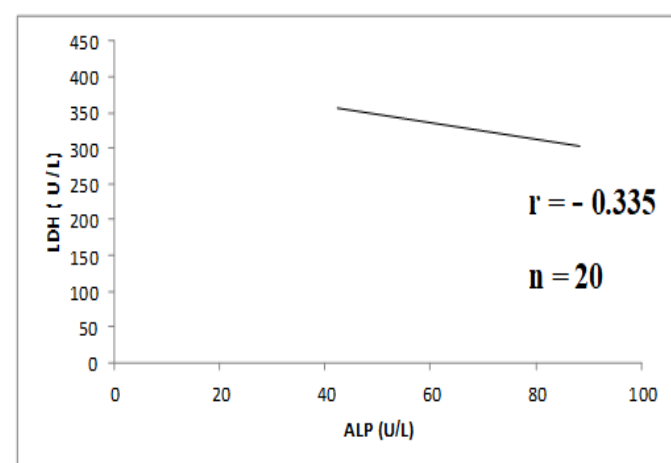


Figure 7 illustrates the relationship between ALP and LDH activity in fertile individuals' seminal plasma.

In the seminal plasma of fertile males ($r = 0.242$) and normospermia ($r = 0.057$), the results showed positive associations between AST and LDH activity (Figs. 8 & 9).

4- Discussion

A significant societal and marital issue that is becoming more commonplace globally is infertility. Infertility is defined as a couple's reduced or nonexistent ability to procreate; subfertility is a word that excludes sterility, which is the whole inability to bear children. Following the study of seminal fluid, the World Health Organization (WHO) calculated the infertility patients by examining the semen parameters (11).

The primary component needed for spermatozoa's metabolic process, as well as for sperm survival, function, and carriage in the female genital canal, is seminal plasma (12), The majority of the ejaculate volume comes from accessory glands, such as the seminal vesicle, prostate, and Cowper's glands, which provide the majority of the ejaculate volume. Small quantities come from the testis and epididymis secretions. The majority of seminal fluid is made up of the secretion from seminal vesicles (5).

ALP enzyme secretion from the testis and prostate has been linked to human research; the source and amount of ALP enzyme in the seminal fluid vary according to the species (13). ALP function in the seminal plasma may have been indicated by the non-significant ($P < 0.05$) variations in seminal plasma ALP activity between normospermic and fertile males (Fig. 1). The activation of fructose precursors to provide amounts of fructose may suit the sperm count or may refer to the compatibility of the sperm count with the ALP secretion from

the prostate and testis glands, according to a clinical study that notes that the ALP enzyme reacts with fructose precursors released in the accessory gland (14).

ALP is an enzyme that is attached to the PI position on the sperm surface and is responsible for keeping the sperm quiet and preventing premature capacitation, which could weaken the ability of fertilization. A limited number of searches have been conducted on the potential role of ALP in sperm function, depending on the concentration of sperm (15). As spermatozoa travel through the female genital canal, they get free of seminal plasma proteins and other covering elements that de-capacitate them, go through the process of capacitation, and develop the capacity to fertilize (16).

Additionally, depending on the sperm count required for these components, the results showed non-significant differences ($P>0.05$) in seminal plasma AST activity between the fertile and normospermia groups (Fig.2). This could indicate the normal activity of gland secretion or the role of the AST enzyme in providing the equilibrium of semen components. This enzyme has been studied for the integrity of the sperm membrane in several studies, but its presence in the seminal plasma has received less attention. In their study of this enzyme between seminal plasma and serum, other study (17), reported that the AST activity in the seminal plasma was 13.71 times higher than that of the serum, whereas another research (18) noted the important relationship between AST enzymatic activity and rate of morphologically abnormal spermatozoa.

Mammalian seminal plasma contains high glutamate concentration, the role of glutamate does not seem to constitute a significant energy source, and some amino acids are beneficial to spermatozoa during storage (19). Aspartate-amino transaminase enzyme catalyzes the transfer of glutamate amino group to oxaloacetate which eventually results in the formation of aspartate and α -ketoglutarate respectively in reverse reaction (20).

The LDH activity for the groups of men with normospermia and fertile men did not differ statistically ($P>0.05$), this finding might be related to the normal activity of prostate secretion or the similarities in semen parameters between the groups of fertile and normospermia as well as the correlations between LDH activity and sperm parameters. LDH activity in the seminal fluid increased in tandem with the percentage of live and normal sperm. According to (21), LDH is said to have a significant metabolic function in sperm capacitation and fertilization, or maybe referred to prostate function as the main source, which may indicate the role of the enzyme in finding the balance between the pyruvate and lactate, these components may be important in the buffering competence of semen or may be referred to block out in the gland duct because of inflammation or bacterial infection. Pyruvate and lactate are present at high concentrations in oviductal fluid, and hence they are commonly utilized as energy substrates by mammalian spermatozoa (22).

4-1 The correlations between enzymes:

4-1 -1 The correlations between enzymes ALP and AST

Alkaline phosphatase is important in the dephosphorylation of

glucose-6-phosphate; however, fructose seems to be synthesized in seminal vesicles whereas ALP is of testicular-epididymal source. ALP reacts with fructose precursor released in the accessory glands. The activation of fructose precursors by ALP could be one of the triggers for spermatozoa activation when they come in contact with seminal plasma. Aspartate amino-transferase catalysis is the reverse conversion of L-glutamate and oxaloacetate to aspartate and α -ketoglutarate (23).

The results revealed a negative correlation between ALP and AST activities in the seminal plasma of fertile and normospermia men (Fig 4 & 5), which may refer to the function of these enzymes in the seminal plasma to provide and save the homeostatic system between nutrients and amino acid or may be referred to the variation of the components in the seminal plasma of these group depend on the sperm concentration and prostate secretions. The testicular-epididymal origin for ALP has been previously confirmed. The ALP enzyme was not found in semen samples of vasectomized boars (24).

4-1 -2 The correlations between enzymes ALP and LDH

New research (25) mention the ALP enzyme keeps spermatozoa inert until they are ejaculated and modulates the acquisition of the fertilizing ability, also LDH is important in sperm capacitation, fertilization, and metabolism.

The results in the present study showed a positive correlation in normospermia (Fig 6) when compared to the fertile group which has a negative correlation (Fig 7), which may refer to the functions of these enzymes in the seminal plasma, and how regulation of the semen components (fructose, lactate, and pyruvate) provide energy required for capacitation and fertilization, or may be refer to the response of several patients in these groups to treatments that regulated the glands secretions to preamble the appropriate conditions for spermatogenesis and maintain this process. Because very little information is available about the correlations between sperm parameters and these enzyme activities of seminal plasma, the important correlations may reflect the roles of these enzymes in the female reproductive tract, which may make sperm safe against highly viscous mucus or during an immune attack, because the female reproductive canals in several mammals secreted a large amount of mucus (23).

4-1-3 The correlations between enzymes AST and LDH:

The estimation of the biochemical components and enzymes in seminal plasma and the correlation between them can be suggested or indicated as biological markers for seminal quality since their levels or values indicate sperm function, integrity, and damage (26). Other study pointed out the significant positive correlation ($P<0.05$) ($r=0.66$) between LDH and AST activity in Jack's semen after the hypoosmotic test, also new research observed a positive correlation between these enzymes in the ram semen and refer to every increase in the percentage of live and normal sperm corresponded to an increase in LDH activity in the seminal fluid (27).

The results showed positive correlations between AST and LDH activities in the seminal plasma of fertile and

normospermic men (Fig. 8 & 9), which may refer to the sharing of these enzymes in the source (prostate), or the function of these enzymes in the energy saving that depend on the sperm concentration. The correlations in study groups may be referred to the effect of treatment response such as testimony is used as testosterone replacement therapy in male hypogonadal disorders and an effective steroid which gives the user high-quality muscles and body strength gains, and who causes several problems in the function of the sex accessory gland (28), or maybe refer to the correlation of these enzymes with the oxidative stress, recent research concluded the higher oxidative stress associated with poorer semen quality and decreased antioxidant capacity in semen (29). These negative effects might be a result of decreased activities of antioxidant enzymes that lead to sperm membrane necrosis as a result of oxidative stress and enzyme liberation, or the correlations between these enzymes may be normal because important in the hyperactivation or capacitation in the female reproductive system (30).

5. Conclusion

It was concluded that the importance of ALP, AST, and LDH activity to detect the capability of gland secretion and normal semen components. There are no significant differences between normospermia and fertile men in the ALP, AST, and LDH activity levels.

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