

## **Study the defects of mitochondrial architecture in mid piece of sperms in young and aged local bulls**

### **دراسة التشوهات في تركيب مايتوكوندريا القطعة الوسطية للحيامن في الثيران المحليه الصغيرة والمسنة**

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#### **Abstract**

This study was conducted in kerbala, vet. Med.lab\ University of kerbala, to determine the effect of age on sperm activity and quality in bulls.

The study included the collection of 30 semen samples from the tail of epididymis from local healthy bulls (15 from young and 15 from aged bulls). The results revealed abnormal mid piece of sperms in 4 (26.66%) samples in young bulls, gives 5 defects as the following: 3 (20%) thickness, 1(6.66%) irregular, and 1(6.66%) absent) in young samples, while the abnormalities of aged shows 13(86.66%) samples gives 42 defects as the following: 10 (66.66%) thickness, 8 (53.33%) interrupted, 6 (40%) irregular, 5 (33.33%) thin, 4 (26.66%) short, 4 (26.66%) pseudo droplet, 3 (20%) absent, 2 (13.33%) double.

Statistical analysis revealed, there is significant variation  $P \leq 0.01$  in abnormalities of sperm mid piece between young and aged bulls.

#### **المستخلص**

أجريت الدراسة الحاليه في محافظة كربلاء المقدسه - مختبر كلية الطب البيطري/جامعة كربلاء لغرض تحديد تأثير العمر على نوعية وكفاءة الحيامن في الثيران. تضمنت الدراسة جمع 30 نموذج للسائل المنوي من ذيل البربخ لثيران محليه ذات صحه جيده (15 من ثيران صغيرة العمر و15 من ثيران كبيرة العمر). بينت النتائج وجود حالات عيوب في القطعه الوسطيه للحيامن في 4(26.66%) حالات من الثيران الصغيرة العمر. إذ أظهرت وجود 5 تشوهات موزعه كالآتي: 3(20%) تتخن, 1(6.66%) تقطع, وحالة واحده (6.66%) اختفاء بالقطعه الوسطيه في نماذج الثيران الصغيره. اما العيوب في الثيران الكبيره اظهرت 13(86.66%) حالة وكانت على 42 تشوه موزعه كالآتي: 10(66.66%) تتخن, 8(53.33%) تقطع, 6(40%) عدم انتظام, 5(33.33%) رفيع, 4(26.66%) قصر, 4(26.66%) قطيره كاذبه, 3(20%) اختفاء, 2(13.33%) ازدواج القطعه الوسطيه.

اظهر التحليل الاحصائي وجود اختلاف معنوي ( $P \leq 0.01$ ) في نسبة تشوهات القطعه الوسطيه للحيامن بين الثيران الصغيره والمسنة.

#### **Introduction**

The fertility of the spermatozoa is directly correlated with the percent motility of spermatozoa. (1) Recommended not to tolerate  $>20\%$  abnormalities of sperm head and / or mid piece in routine AI practice.

Spermatozoa motility is the sperms ability to move, the movement is slow, not in straight line, the spermatozoa face difficulty in pass through the cervical mucus and penetrating the hard outer shell of the egg (2). Total sperm abnormalities were positively significant influenced by abnormalities at head (0.650) and mid-piece (0.723) regions (3). Mitochondria are the organelles responsible for producing the majority of a cell's adenosine triphosphate (ATP) and also play an essential role in gamete maturation and embryo development(4). ATP production within the mitochondria is dependent on proteins encoded by both the nuclear and the mitochondrial genomes, therefore co-ordination between the two genomes is vital for cell survival (5).

Considerable attention has been paid to the effect of age on sperm morphology of bulls (6) (7) (8). The ratio between the total forms of abnormality in the head, mid piece and tail of spermatozoa was as 1:1.5:1. (9)

Therefore this study included:

- 1- Semen collection from tail of epididymis in bulls.
- 2- Sperms stain by eosin and ethidium bromide
- 3- Florescent and light microscopical examination.

**Materials and methods**

Thirty semen samples were collected from tail of epididymis in healthy bulls, used needle gage 23. Fifteen samples were collected from young bulls(2-4 years) and 15 samples were collected from aged bulls (over6years),in order to detected the abnormalities of mid piece of sperms.

Sperms were stained with eosin, and ethidium bromide dyes to examination by UV light. The samples were subjected to florescent and light microscopical examination, in order to investigation the defects of mid piece with the defect in the mitochondrial structure.

The data statistically analysis by SAS program (2001) (10).

**Results**

Thirty semen samples subjected to microscopical examination gives the results , **15 samples** from young bulls (**table 1**) appeared 4 (36.66%) samples abnormal mid piece, gives 5 mitochondrial defects in mid piece as the following: 3(20%) thickness, 1(6.66%) irregular, and 1(6.66%) absent; while the defects of aged bulls (**table 2**) appeared 13( 86.66%) samples abnormal mid piece, gives 42 defects as the following : 10 (66.66 %) thickness, 8 (53.33%) interrupted, 6 (40%) irregular, 5 (33.33%) thin, 4 (26.66%) short, 4 (26.66%) pseudo droplet, 3 (20%) absent, 2 (13.33%) double.

The total defects in young bull reached (4) 36.66% while the defects in aged reached (13) 86.66%, (**Table 3**)

Statistical analysis revealed,there is significant variation  $P \leq 0.01$  in abnormalities of sperm mid piece between young and aged bulls.

Table 1: Show in details the defect in the sperm in young bulls

No	interrupted	thickness	irregular	Thin	short	double	Pseudo-droplet	absent
1								
2								+
3								
4								
5		+						
6								
7								
8		+						
9								
10		+	+					
11								
12								
13								
14								
15								

Table 2 Show in details the defects in the sperm in aged bulls

No	interrupted	thickness	irregular	Thin	short	double	Pseudo-droplet	absent
1	+	+	+					
2	+	+					+	
3	+	+		+		+		
4								
5		+	+		+			+
6	+			+		+		
7	+	+						
8			+	+	+			
9	+	+					+	+
10		+						
11	+	+	+				+	
12				+	+			+
13								
14	+	+	+	+				
15		+	+		+		+	

Table 3: Show the total normal and defect of sperms with the percent

	Young bulls	Aged bulls	Total
Normal	11 (73.33%)A	2 (13.33%)B	13 (43.33%)
Defect	4 (36.66%) B	13 (86.66%)A	17 (56.66%)

The rates which have different letters within same raw are significant variation  $P \leq 0.01$

## Discussion

We are utilized the florescent microscope to investigation of mitochondrial defect to visualized the genomic structural. (11) Mention, the phase contrast and fluorescent microscopy of semen samples showed large numbers of spermatozoa with short, rigid, thick and irregular tails.

In another stain, the eosin used to appear the abnormalities of structural mid piece. This is agreement with (12)

The defect of mid piece in young bulls reached 4 (36.66 %) this result compare with the result of aged bull 13(86.66%) reflex the role of age in the sperm quality. Effect of age was significant (P, 0.01) on most of the semen characteristics and spermatozoal morphology (13)

The results of this study are in agreement with that of (14), (15), and (16) in that the variation in the percentage of normal sperm and the frequency of abnormal sperm increased with ageing.

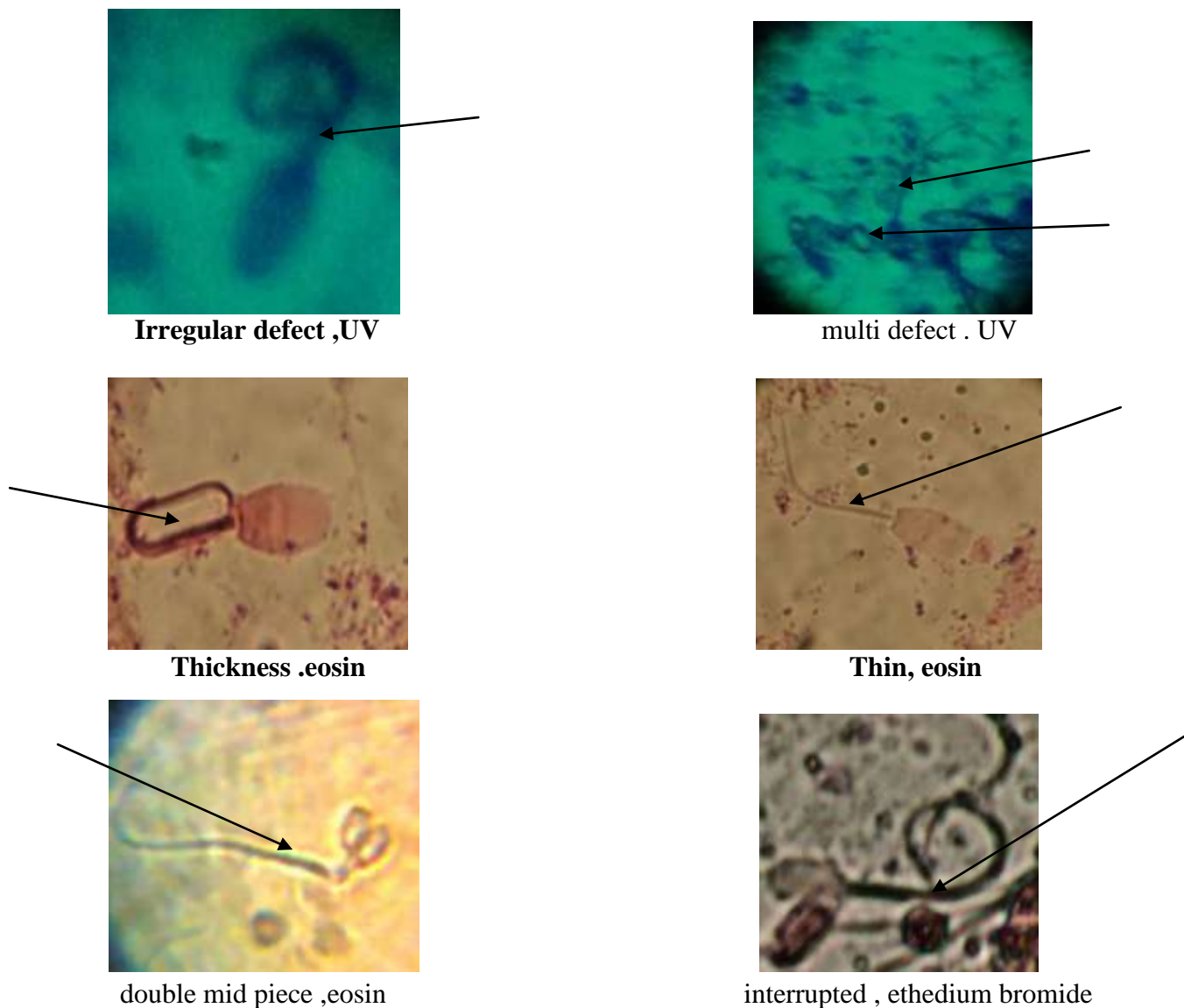


Figure 1: show the defects in the mid piece of sperms .stained with eosin, and ethidium bromide, exam, by light and florescent microscope

While the results are in contrary to the results obtained by (17), (18), (19), and (20) reported that the morphology of spermatozoa, sperm concentration, semen motility and the volume of the ejaculate improve with an increase in the age of bulls.

Recent studies have shown that 11% or more head, mid-piece or tail abnormalities and 18% or more total abnormalities of spermatozoa are associated with reduced fertility in bulls (21).

Semen samples of young bulls appear 4 (36.66%) of mid piece abnormalities gives 5 different types of defects: 3 (20%) thickness, 1(6.66%) irregular, and 1(6.66%) absent. While the abnormalities in the aged bulls 13(86.66%) gives 52 defects: 10 (66.66 %) thickness, 8 (53.33%) interrupted, 6 (40%) irregular, 5 (33.33%) thin, 4 (26.66%) short, 4 (26.66%) pseudo droplet, 3 (20%) absent, 2 (13.33%) double.

Major sperm defects that were affected by age included pyriform, knobbed acrosomes, mid-piece reflexes and dag defects (22).

Significant proportions of head and tail abnormalities were observed in bulls belonging to different age groups. The highest percentage of total tail abnormalities was recorded in 6-<8 yrs (23).

One of the important indications of sperm viability, Semen volume, concentration of spermatozoa, and motility of spermatozoa are recognized as important indices of semen quality (24). The defect of mitochondria lead to low motility of sperm because the mitochondria responsible for energy of sperm (25) therefore the study concentrated of the mitochondrial defect, in order to give the important of this part of sperm viability.

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