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Effects of raw *Aloe barbadensis* leaf gel on quality and fertilising potential of extended semen from Red Sokoto bucks

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Abstract

Semen evaluation is a routine practice in breeding soundness evaluation and investigation of fertility breeder male farm animals. Artificial insemination (A.I.) requires an objective and rapid, but inexpensive, method to evaluate ejaculates. The goal of semen assessment is to predict fertilising ability of the semen of breeding buck. A study was conducted to investigate the effects of raw Aloe barbadensis leaf gel (Gel_{raw}) on quality and spermatozoa fertilising potential of extended goat semen. Semen was collected from buck using electro-ejaculator and were diluted with raw Aloe barbadensis Gel (Gel_{raw}) at 7.00(GEYCE)+0.00(Gel_{raw}), 5.25(GEYCE)+1.75(Gel_{raw}), 3.50(GEYCE)+3.50(Gel_{raw}), 1.75(GEYCE)+5.25(Gel_{raw}), 0.00(GEYCE)+7.00(Gel_{raw}) as T1, T2, T3, T4 and T5 respectively. Extended semen samples were evaluated at 0, 24, 48 and 72 h for quality and spermatozoa fertilising potential. All treatments were replicated thrice in completely randomized design. At 72-h, Normal Spermatozoa (NS) and Acrosome Integrity (AcI) of 5.25g/L Gel_{raw} treated samples significantly increased to 78.33±2.0% and 82.23±4.3% compared to 67.33±7.0% and 73.33±5.0% respectively in the control while ProgressiveMotility (PM) and Plasma Membrane Integrity (PMI) were not significantly affected. Aloe barbadensis gel (Gel_{raw}) added to diluent at 7.00g/L offered cytoprotection to extended buck spermatozoa for 72 hours.

Keywords: Artificial insemination, buck semen, *Aloe barbadensis* leaf gel, frozen-thawed semen

Effets du gel brut de feuilles d'*Aloe barbadensis* sur la qualité et le potentiel fertilisant du sperme étendu de boucs Red Sokoto

Résumé

L'évaluation du sperme est une pratique courante dans l'évaluation de la solidité de la reproduction et l'étude de la fertilité des animaux de ferme mâles reproducteurs. L'insémination artificielle (IA) nécessite une méthode objective et rapide, mais peu coûteuse, pour évaluer les éjaculats. L'objectif de l'évaluation de la semence est de prédire la capacité fécondante de la semence des boucs reproducteurs. Une étude a été menée pour étudier les effets du gel brut de feuilles d'Aloe barbadensis (Gel_{raw}) sur la qualité et le potentiel fertilisant des spermatozoïdes du sperme de chèvre étendu. Le sperme a été collecté sur des boucs à l'aide d'un électro-éjaculateur et dilué avec du gel brut d'Aloe barbadensis (Gel_{raw}) à 7,00 (GEYCE) + 0,00 (Gel_{raw}), 5,25 (GEYCE) + 1,75 (Gel_{raw}), 3,50 (GEYCE) + 3,50 (Gel_{raw}). , 1,75(GEYCE)+5,25(Gel_{raw}), 0,00(GEYCE)+7,00 (Gel_{raw}) comme T1, T2, T3, T4 et T5 respectivement. Des échantillons de sperme étendus ont été évalués à 0, 24, 48 et 72 h pour déterminer leur qualité et leur potentiel fécondant les spermatozoïdes. Tous les traitements ont été répétés trois fois selon une conception complètement randomisée. Après 72 heures, l'intégrité normale des spermatozoïdes (NS) et de l'intégrité des acrosomes (IAc) des échantillons traités à 5,25 g/L de Gel_{raw} a augmenté de manière significative à 78,33 ± 2,0 % et 82,23 ± 4,3 %, contre 67,33 ± 7,0 % et 73,33 ± 5,0 % respectivement dans le contrôle tandis que la motilité progressive (MP) et l'intégrité de la membrane plasmatique (IMP) n'étaient pas affectées de manière significative. Le gel d'Aloe barbadensis (Gel_{raw}) ajouté au diluant à raison de 7,00 g/L a offert une cytoprotection aux spermatozoïdes de chevreuil prolongés pendant 72 heures.

Mots-clés: Insémination artificielle, sperme de chevreuil, gel de feuilles d'*Aloe barbadensis*, sperme congelé-dégelé

Introduction

Artificial insemination is a crucial technology for livestock producers world-wide and this technology has afforded producers access to premier genetics without having to physically own superior sires Dalen *et al*., 2021. Most goat farmers do not use any standard breeding programme at this moment. However, there are few individuals with good conformation and qualities among the local breeds, but this population is undefined and unselected (Dubeuf and Boyazoglu, 2009). The success of AI depends on how the ejaculate is handled, stored and inseminated (Leboeuf *et al*., 2000). For ejaculate that will be used shortly after collection or that is to be used in a small flock size, it is best used fresh or chilled. Nonetheless, frozen-thawed semen has an advantage over fresh ejaculate as it can stored for a longer period and shipped to anywhere in the world for use. Unfortunately, during the cryopreservation process, the spermatozoon suffers structural damage, reducing its viability and fertilizing potential compared to fresh or refrigerated sperm (Salamon and Maxwell, 2000). The presence of seminal plasma, the type and concentration of cryoprotectant used, and the preservation procedure, all plays important role in the structural damage during cryopreservation (Watson, 2000; Purdy, 2006).

The mode of operation of cyto-preservative in stabilising and protecting cells while suppressing the formation of ice crystal is not fully known. Understanding of this process will improve freezing protocols and formulating efficient cyto-preservative solutions. Glycerol is miscible with water molecules hence hindering the formation of an extended water network in the solution and hence the formation of ice crystals (Towey *et al.,* 2011). The surface penetrating ability of *Aloe vera* extract and its solubility in intracellular water could offer an excellent solution which could prevent ice crystal formation in the semen hence; this study is directed towards the possibility of on farm replacement of glycerol with Aloe vera extracts which could be easily assessable for on farm preservation of semen. Aloe vera has been classified as medicinal plant with specific bio-active ingredients such as alkaloids, saponins, flavonoids, proteins, lipids, amino acids, vitamins C, B (1, 2 and 6), A, E, enzymes, organic and inorganic compounds, and mineral salts such as sodium, calcium, iron, potassium, chloride, manganese, copper, and zinc (Hamman, 2008; Saeedeh *et al*, 2021). It has been used as a natural antioxidant with high potential of reducing fat oxidation and oxidative stresses (Vinson *et al.,* 2005). Also, pharmacological effects of Aloe vera viz: anti-inflammatory, anti-arthritic, and antimicrobial have been largely documented (Newall *et al*., 1996) but very few studies on effects of the leaf extracts on spermatozoa characteristics have been reported.

Materials and Methods

Experimental site

Semen collection by electro-ejaculator was done at the Small Ruminant Unit of the University of Ibadan Teaching and Research Farm while the semen quality and spermatozoa fertilizing potential were evaluated at the Animal Physiology and Biotechnology Laboratories, Department of Animal Science, University of Ibadan, Ibadan, Oyo State Nigeria ($7^{o}20^{l}$N, $3^{o}50^{l}$E; 200m above sea level).

Preparation of raw Aloe barbadensis leaf gel

Freshly harvested and authenticated *Aloe Vera* leaves were washed; the outer layers carefully peeled with forceps to prevent staining of the colourless viscous inner gel with chlorophyll. The gel was carefully scooped with a spatula into a beaker, homogenised and covered with foil paper and kept at the temperature of 37 $^{\circ}$C in a water bath prior to use. This product was designated Gel_{raw}.

Semen collection

Three matured and healthy Red Sokoto bucks aged 2-3 years, weighing (41.5 ± 2.0 kg) were used. They were ejaculated weekly using electro-ejaculation method as described by Oyeyemi *et al*., (2000).

Semen extender and experimental treatments layout

Presented in Table 1 is the composition of the glucose-egg yolk-citrate extender (GEYCE) used for the experiment. The mixture was centrifuged and the supernatant decanted into a universal bottle and the pH adjusted to 7 using a buffer solution. The diluent was kept in the refrigerator (-1 ˚C) until required for use. Five experimental extenders were formulated by adding Gel_{raw} at 0.00, 1.75, 3.50, 5.25 and 7.00 mL in the extender representing 0, 25, 50, 75 and 100 % replacement for glycerol as a cyto-preservative. The experimental layout is presented in Table 1.

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Glucose-egg yolk-sodium citrate extender

Component	Quantity
Sodium Citrate (g)	2.37
Egg yolk (mL)	18.00
Glucose (g)	0.80
Penicillin (g)	0.06
Streptomycin (g)	0.10
Glycerol (mL)	7.00
Distilled Water (mL)	78.70

Source: Modified from Chaudhari and Mshelia; (2002).

Table 1: Experimental layout (Gel_{raw})

Treatment (Gel_{raw}, g/L)	Composition
T_1	7.00 (GEYCE) + 0.00 (Gel_{raw})
T_2	5.25 (GEYCE) + 1.75 (Ge^{l}_{raw})
T_3	3.50 (GEYCE) + 3.50 (Gel_{raw})
T_4	1.75 (GEYCE) + 5.25 (Gel_{raw})
T_5	0.00 (GEYCE) + 7.00 (Gel_{raw})

Gel_{raw} = Raw *Aloe barbadensis* gel; GEYCE = glucose-egg yolk-citrate extender

Pre-extension evaluation of semen

The semen in collection tube was immersed in warm water at 37 °C before evaluation. The sample was observed for consistency, colour, volume, and mass activity, normal spermatozoa (NS) and spermatozoa concentration (SC). The ejaculate volume was estimated by reading the meniscus of the calibrated tube. Colour was assessed by visual observation. The consistency, mass activity and progressive motility (PM) were determined using methods described by Chenoweth, 2005.

Semen processing

Semen samples were diluted in part A of each extender at 37 °C. Diluted semen and the part B of the extenders were gradually cooled to 4°C in the refrigerator before addition of part B extender. The addition of part B extender was carried out in three steps at 30 minutes interval for equilibration at this temperature before freezing to -22 °C for evaluation at 0, 24, 48 and 72 hours.

Post-thaw semen quality and spermatozoa fertilising potential evaluation

Semen quality assessment; Plasma Membrane (PM), spermatozoa liveability (SL), NS and pH, and spermatozoa fertilising potential assessment; plasma membrane integrity (PMI) and acrosome integrity (AcI) were carried out at 0, 24, 48 and 72 hours. Spermatozoa progressive motility was assessed with a phase contrast microscope at x400 (37 °C). Normal spermatozoa and SL were assessed from two hundred spermatozoa stained with nigrosin–eosin stain per sample reading (Maxwell, 1987). The AcI percentage was estimated from smears stained with nigrosin–eosin examined under phase contrast microscope at x1000 magnification under oil immersion objective and bright field (Yildiz *et al*., 2000). A total of 200 spermatozoa in four microscopic

Table 2: Characteristics of collected fresh buck semen

Characteristics	Value
Mass Activity	+++
Semen Volume (mL)	0.75 ± 0.24
pH	6.60 ± 0.18
Progressive Motility (%)	95.00 ± 2.06
Spermatozoa Concentration ($\times 10^6$/mL)	667.00 ± 37.56
Spermatozoa Liveability (%)	99.00 ± 1.50
Normal Spermatozoa (%)	98.00 ± 2.25
Acrosome Integrity (%)	98.00 ± 3.19

fields were counted. The PMI was assessed by hypo-osmotic swelling test (Buckett *et al*., 1997).

Statistical analyses

Data collected were subjected to descriptive statistics and one way Analysis of variance procedure of SAS, (2011) and means were compared using Duncan's multiple range test of the same software.

Results

+++ = Very strong wave motion

Table 3: Shows the mean characteristics of fresh ejaculate of red Sokoto goat. All parameters evaluated indicated that the samples were of high quality with high spermatozoa fertilising potential.

Table 4: Effect of Gel_{raw} on quality and spermatozoa fertilising potential in extended (-22 °C) buck semen at 0 hour

Parameters	Gel_{raw} (g/L)					
	0.00	1.75	3.50	5.25	7.00	SEM±
Progressive Motility (%)	78.33^{a}	66.67^{b}	63.33^{b}	73.33^{a}	73.33^{a}	1.72
Spermatozoa Livability (%)	98.00^{a}	95.67^{a}	97.00^{a}	93.33^{ab}	90.33^{ab}	0.89
Normal Spermatozoa (%)	97.33^{a}	91.33^{b}	95.00^{a}	91.00^{b}	90.67^{b}	0.85
pH	6.43^{ab}	6.33^{b}	6.33^{b}	6.57^{a}	6.37^{ab}	0.03
Acrosome Integrity (%)	97.33	97.67	98.00	97.67	96.67	0.15
Plasma Membrane Integrity (%)	96.67	93.33	95.00	96.67	91.67	0.63

^{ab} =In a row, values having different alphabet differ statistically (P<0.05), SEM is Standard Error of Means.

Presented in Table 4 are semen quality assessments at 0 hour of extension. The effect of increasing Gel_{raw} concentrations on AcI and PMI is not significant (p>0.05). There were statistical (p<0.05) variations in PM, SL, NS, and pH. However, values for all parameters were within acceptable range of values for high quality semen.

Table 5: Effect of Gel_{raw} on quality and spermatozoa fertilising potential in extended (-22 °C) buck semen at 24 hours

Parameters	Gel_{raw} (g/L)					
	0.00	1.75	3.50	5.25	7.00	SEM±
Progressive Motility (%)	76.67^{a}	68.33^{b}	68.33^{b}	65.00^{b}	68.33^{b}	1.23
Spermatozoa Livability (%)	85.67	89.33	85.00	87.67	86.00	0.51
Normal Spermatozoa (%)	80.67^{ab}	89.33^{a}	82.00^{ab}	76.33^{b}	87.00^{a}	0.50
pH	6.87^{a}	6.80^{b}	6.80^{b}	6.80^{b}	6.80^{b}	0.01
Acrosome Integrity (%)	84.33^{b}	91.33^{a}	82.67^{b}	77.67^{c}	83.33^{b}	1.48
Plasma Membrane Integrity (%)	86.67^{a}	80.00^{ab}	76.67^{b}	83.33^{ab}	63.33^{c}	2.60

^{ab} = In a row, values having different alphabet differ statistically (P<0.05), SEM is Standard Error of Means

Effect of Gel_{raw} on quality and spermatozoa fertilising potential in extended (-22 °C) buck semen at 24 hours

At 24 hours post extension and -22 °C storage temperature (Table 5) PM was considerably ($p<0.05$) higher in samples extended with the control than in Gel_{raw} containing extender. Normal spermatozoa in sample extended with 0.00 g/L of Gel_{raw} were similar ($p>0.05$) to those extended in Gel_{raw}. However, samples extended with 1.75 g/L of Gel_{raw} were 13 % higher than those extended in 5.25 g/L $Gel_{raw.}$ Again, samples in 0.00 g/L of Gel_{raw} were 0.07 % higher than all samples extended in $Gel_{raw.}$ Samples preserved in 1.75 and 5.25 g/L of Gel_{raw} has 7 % and 6.66 % higher number of spermatozoa with intact acrosome. However, 10 % and 23.34 % of samples in the 0.00 g/L of Gel_{raw} has higher number of spermatozoa with intact plasma membrane compared to those in 3.50 and 7.00 g/L of Gel_{raw} respectively

Table 6: Effect of Gel_{raw} on quality and spermatozoa fertilising potential in extended (-22 °C) buck semen at 48 hours

Parameters	Gel_{raw} (g/L)					
	0.00	1.75	3.50	5.25	7.00	SEM ±
Progressive Motility (%)	56.67^{c}	70.00^{a}	63.33^{b}	63.33^{b}	61.67^{b}	1.38
Spermatozoa Liveability (%)	74.00^{b}	77.33^{ab}	80.33^{a}	80.67^{a}	83.67^{a}	1.06
Normal Spermatozoa (%)	71.00^{c}	80.00^{b}	74.00^{bc}	78.00^{b}	86.00^{a}	1.66
pH	6.87	6.87	6.90	6.87	6.87	0.01
Acrosome Integrity (%)	73.67	71.67	77.33	75.00	77.67	0.73
Plasma Membrane Integrity (%)	43.33^{b}	53.33^{a}	50.00^{a}	53.33^{a}	33.33^{c}	0.45

^{abc} = In a row, values having different alphabet differ statistically ($P<0.05$), SEM is Standard Error of Means

Effect of Gel_{raw} on Quality and Spermatozoa Fertilising Potential in Extended (-22 °C) Buck Semen at 48 Hours

In Table 6, there were significantly ($p<0.05$) higher values obtained for PM, SL, NS and PMI for all semen samples extended with Gel_{raw} at various concentration after 48 hours post extension and storage compared to samples extended with 0.00 g/L of Gel_{raw}. However, while samples extended with 1.75 g/L of Gel_{raw} has higher percentage of PM and PMI, samples in 7.00 g/L of Gel_{raw} extender had higher proportion of LS and NS. There was no significant ($p>0.05$) variation in results obtained for semen pH and AcI.

Table 7: Effect of Gel_{raw} on quality and spermatozoa fertilising potential in extended (-22 °C) buck semen at 72 hours

Parameters	Gel_{raw} (g/L)					
	0.00	1.75	3.50	5.25	7.00	SEM±
Progressive Motility (%)	48.00^{ab}	50.00^{ab}	58.33^{a}	56.67^{a}	51.67^{ab}	1.27
Spermatozoa Liveability (%)	72.67^{b}	87.33^{a}	70.00^{b}	80.67^{ab}	77.67^{ab}	1.97
Normal Spermatozoa (%)	67.33^{b}	77.33^{a}	79.33^{a}	78.33^{a}	77.33^{a}	1.41
pH	6.87	6.87	6.90	6.87	6.87	0.02
Acrosome Integrity (%)	73.33^{b}	80.00^{a}	70.33^{b}	82.33^{a}	79.67^{a}	1.46
Plasma Membrane Integrity (%)	56.67^{a}	56.67^{a}	60.67^{a}	61.67^{a}	50.00^{ab}	1.33

^{ab} = In a row, values having different alphabet differ statistically ($P<0.05$), SEM is Standard Error of Means.

Effect of Gel_{raw} on quality and spermatozoa fertilising potential in extended (-22 °C) buck semen at 72 hours

At 72 hours of storage PM were ≥ 50 % in all samples extended in extender with *Aloe vera* gel but lower than 50% in extender with 0.00 g/L of Gel_{raw}. Although the values were not statistically ($p>0.05$) significant, however, values recorded for samples in 3.50 g/L of Gel_{raw} was 10.33 % higher than the control. For all treatments, the PMI results were statistically ($p>0.05$) equivalent including the control but there were statistical ($p<0.05$) variations in values obtained for SL, NS, AcI, and semen pH (Table 7). Spermatozoa liveability was 14.66 % higher in samples extended with 1.75 g/L of $Gel_{raw.}$ Samples in 3.50 g/L of Gel_{raw} have 12 % higher NS compared to samples in 0.00 g/L of $Gel_{raw.}$ The values for PMI were statistically similar ($p>0.05$) for all treatments except for samples in 7.00 g/L of Gel_{raw} which was significantly lower than the rest. All of the samples' SAI were statistically ($p<0.05$) greater than those 7.00 g/L of Gel_{raw}, with the exception of the sample extended with 0.00 g/L of Gel_{raw}, which was statistically ($p>0.05$) equivalent to the control. However, the values were still within the recommended range for buck semen.

Discussion

There was reduction in parameters evaluated for semen diluted in extenders containing different concentrations of Gel_{raw} and the control compared to fresh semen fresh ejaculate but the reduction was not significant for PM, AcI and PMI. However, the reduction was significant for SL, NS, and pH. Nevertheless, the values obtained for all parameters are high enough to achieve satisfactory results if inseminated. Motility is commonly used for the assessment of viability of spermatozoa in an ejaculate prior to, and after the preservation process (Johnson *et al*. 2000). Values obtained in this study for PM declined from 95 % in the undiluted sample to a range of 63.3 ± 2.9 to 73.3 ± 5.8 % in samples extended in extenders containing Gel_{raw} and 78.3 ± 17.6 % for the control. Results obtained in this study shows that the concentration of Gel_{raw} in the samples used for the experiment did not significantly affect PM as reported by Sundararaman and Edwin (2008). There was no significant difference in the SL of the control and samples extended with Gel_{raw} except in sample containing 7 00 g/L which is significantly lower than the control. However, the value of SL (90.3 ± 4.7 %) is higher than values reported by Okukpe *et al*., (2012) when CUE, whole milk, soymilk and soymilk-CUE extenders were used to extend ram semen. The NS decreased from 98 ± 1.2 % in fresh semen to 97.3 ± 1.0 % in the control and between 90.67 ± 1.2 % to 95.00 ± 1.0 % in samples extended with varying concentration of Gel_{raw} pre-storage. The reduction was significant between the control and samples extended with Gel_{raw} expect those diluted with extender containing 3.50 g/L. The result shows that less than 10 % spermatozoa in the extended semen samples in all the treatments were morphologically abnormal. Batista *et al*., (2009) obtained morphological abnormality of 3.1 to 6.5 % for fresh semen samples from Majorera goat. El-Bawab *et al.,* (2015) recorded 9.66 % abnormalities in spermatozoa of semen diluted with different concentrations of chicken low density lipoprotein (LDL) extender pre-storage. Roca *et al*., (2006) and Melissa (2002) recommended semen with 85 % and70 % spermatozoa with normal morphology respectively for use for cryopreservation. This implies that all samples extended with Gel_{raw} are suitable for preservation.

The PMI of semen samples extended with Gel_{raw} ranged from 91.67 ± 2.9 to 96.67 ± 5.8 % which implies that most of the spermatozoa in the samples have intact and functional plasma membrane. This range of the result is higher than 80 % recommended by Roca *et al*. (2006). The AcI in this study shows similar trend. There was no significant difference in the values of AcI for all samples extended with Gel_{raw} and the control.

Values (96.67 ± 2.5 to 98.00 ± 1.0 %) were higher than 85.08 % obtained by Iman *et al.* (2015) in Buck Semen extended with different concentrations of Low-Density Lipoprotein (LDL) pre-freezing. Dziekońska *et al*., (2015) also recorded 91.73 %, 90.45 % and 91.00 % apical acrosome integrity for boar semen diluted in Androhep, BTS and GEDIL extender respectively. The pH of Gel_{raw} extended semen samples ranged from 6.33 to 6.57. This implies that pH value pre-storage was slightly acidic, this confirms the reports of Oyeyemi *et al.* (2001) and Purdy (2006) that buck semen is slightly acidic.

Freezing-thawing process causes loss of viability and functionality of spermatozoa (Purdy, 2006). Temperature reduction causes morphological and physiological alterations in spermatozoa. As the temperature lowers from body temperature to 0 to 5 °C, membrane lipids undergo phase shifts, increasing ion permeability and promoting fusion, resulting in irreversible motility loss, disruption of the acrosome, increased morphological defects, and spermatozoa mortality. In an in vitro investigation using post-thaw bull spermatozoa, Gillan *et al*., (2008) found that fertility is related to motility, morphology, concentration, viability, and acrosomal integrity. Effects of Gel_{raw} on post-thawed quality of extended buck semen at 24 hours of storage under -22^{o}C there were no difference in values obtained for SL and NS respectively. All samples still possess appreciable quality. Though, PM, AcI, PMI, and pH differ significantly, all values recorded are within reported recommendations (Meliss, 2002; Roca *et al*., 2006). Semen samples extended with $\mathbf{Gel_{raw}}$ improved semen characteristics after 48 hours of freezing. Though the improvement was not significant for PM, SL, pH and AcI, it was for NS and PMI. The improvement observed in the qualities of samples preserved with Gel_{raw} can be attributed to the bio-active ingredients that serve as a source of additional nutrient to spermatozoa. Also, tannin and saponin that was present in the extracts are active against microbial activities which could cause deterioration destruction of the spermatozoa. Reduction in PM and viability observed after 48 hours of storage in comparison to undiluted semen samples pre-freezing corroborated the report of Salamon and Maxwell (2000), that reduction in PM and SL could be observed after freezing. The damages observed may also be due to the handling protocol (Watson, 2000; Purdy, 2006). Post-thaw evaluation of buck semen samples stored at -22^{o}C shows significant improvement in SL, NS, pH, and AcI in the samples compared to the control after 72 hours of freezing. Results obtained for PMI were similar across treatments and were higher than 36.46 to 45.50 % and 36.54 to 44.06 % recorded by Jun-He Hu *et al.,* (2013) for Shanbei Cashmere buck frozen in extenders containing glycerol (2 to 10 %) and egg yolk (11 to 16 %) respectively. It is also higher than 49.50 ± 1.4 % obtained by Sundararaman and Edwin (2008) but similar to findings of Muhammed *et al*., (2015) who recorded post-thaw PM of 50 to 60 for buffalo semen extended with 10 to 20 % of duck egg yolk. The PM recorded in this trial is similar to 46.00 to 62.20 % obtained by Daramola *et al.* (2016) when buck semen was frozen in Tris-egg yolk extender supplemented with pineapple juice. The values recorded for AcI and PMI in this study were better than those obtained by Daramola *et al*., (2016) for extenders supplemented with Cucumber juice, orange juice and Pineapple juice. Semen of Red Sokoto goat semen used for this study is more acidic pH 6.60 for fresh ejaculate, pH 6.33 to 6.57 for pre-freezing and pH 6.73 to 6.90 after 72 hours of freezing, compared to pH 7.0 and 7.2 recommended to be most favourable for spermatozoa motility for goat semen (Molinia *et al*., 1994; Purdy, 2006). The causes of slightly acidic nature of observed pH could probably be attributed to the use of Electro-ejaculator which is believed to stimulate the buck to release acidic urine that could contaminate semen and reducing the pH (Tarmizi, *et al*., 2020).

Conclusion

This report revealed that the decline in fertilising potential of buck semen extended with aloe vera leaf extract as shown by a decline in all the semen quality parameters as the period of storage increases. However, spermatozoa motility was up to 60% at 72 hours of storage in semen sample extended with raw leaf extracts of aloe vera. Also, since the raw gels of *Aloe barbadensis* maintained plasma membrane integrity of spermatozoa in extended buck semen above 60 % after 48 hours

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