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Effect of Phosphorous and Phosphorous Plus Nitrogen Application on Nutritive Value and *In situ* Digestion Kinetics of Oat (*Evena sativa*) Grass in Nili Ravi Buffalo Bulls

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NAT designed and conducted research trials, got financial support and analyzed data. AF and MAS helped in analysis and write-up. MIB a graduate student worked at the farm and lab and wrote draft manuscript.

Key words
Phosphorous, Nitrogen, Application, Nutritive value, *In situ* digestion kinetics, Oat grass, Buffalo bulls

ABSTRACT

Current study was undertaken to investigate the effect of different levels of phosphorous (P) and phosphorous plus nitrogen (NP) fertilizer on nutritive value and *in situ* digestion kinetics of oat fodder in Nili Ravi buffalo bulls. Four cannulated buffalo bulls were used for *in situ* screening of oat fodder harvested at 100 days of age sown using different levels of phosphorous and phosphorous plus nitrogen fertilizers. Dry matter (DM) and crude protein (CP) content were higher ($p<0.05$) in oat fodder supplemented with P and NP fertilizer as compared to control. The supplementation of P fertilizer resulted in decreased NDF and hemicellulose and did not show any effect on ADF content of oat fodder. At 48 h of incubation, *in situ* digestibility of DM, CP, NDF, ADF and hemicellulose was greater ($p<0.05$) with medium levels (N20P20 and N25P25) of P and NP fertilizer as compared to control. The extent of DM, CP, NDF, ADF and hemicellulose digestion and rate of degradation was increased in response to P and NP fertilization. However, the lag time of DM, CP, NDF, ADF and hemicellulose of oat fodder was the inverse because it decreased in repose to medium level of P and NP fertilization as compared to control.

INTRODUCTION

Nutritional requirements of ruminants are mainly met through fodder crops, shrubs, agro industrial wastes and grains. In future it is expected that ruminants will be more dependent on forages because readily expending human population will have direct competition with livestock for edible grains (Bulla *et al*., 1997). Even in advance countries like United States where grains are in abundant and in-expensive, major caloric requirement is met through green fodder (Fisher *et al*., 1989). Livestock have always been a very important part of the agricultural system of Pakistan, but lacking good quality fodder has been an ongoing major constraint on production.

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Oats, sorghum, sadabahar (*Sorghum bicolor, Sorghum sudanefe*), maize, guar, cowpeas, mott grass, berseem, lucerne, turnip and mustard are commonly available fodders for livestock in Pakistan. However, poor inputs, low genetics potential of fodder varieties, lack of agriculture research and extension services and orthodox agronomic practices leads to low production per acre and lower the quality of the fodders. These facts forced for immediate remedies to solve the fodder shortage in the country for the improvement in livestock production (Sarwar *et al*., 2002).

The important management practices that affect yield and quality of forage include fertilization of fodder crops and harvesting them at proper stage of maturity. Fertilization of grasses with nitrogen and phosphorous typically increases the yields of fodder and their nutritive value (Messman *et al*., 1991). As supplementation of N results in increased crude protein (CP) and dry matter (DM) of the fodder crops. The improved N supply from fodder results in increased DM intake by ruminants (Sarwar and Nisa, 1999).

Phosphorous has been called the "master mineral" because it is involved in most metabolic pathways, is a key component of adenosine 5-triphosphate (ATP)

which transports chemical energy within cells for energy metabolism, and it is a component of DNA. Next to calcium, phosphorous is the most abundant mineral in the body. Approximately 85% of phosphorous in the body can be found in bones and tissues, and roughly 10% circulates in the bloodstream. The remaining phosphorous can be found in cells and tissues throughout the body. Phosphorous helps filter out waste in the kidneys and contributes to energy production in the body by participating in the breakdown of carbohydrates, protein, and fats. Supplementations of P fertilizer in oat grass increased the yields of fodder as developing seedlings of oat can suffer from P deficiency during early growth (Saarela *et al*., 2003). As P increases the germination and also helps in roots development of crops, so supply of P is critical for higher yields of fodder in early sowing season (Grant *et al*., 2001). The quantity and quality of fodder depends upon variety of fodder, ago-chemical conditions and management factors (Rotz *et al*., 2002).

To improve the quality and quantity of green fodder per hectare, it is essential to determine its fertilizer requirements. Different varieties might respond differently to fertilizer application under changing soil and environmental conditions. The plant nutrition may not only affect the forage production but also improve the quality of forage from view point of its protein contents. (Mahmud *et al*., 2003). Oats are important winter fodder in both irrigated and rain fed areas of Pakistan (Dost, 1997). The ideal oat grass should have higher CP and digestibility and low in crude fiber (CF). Improvements in forage utilization are predicted on accurate measurement of the nutritional value of forages.

The objective of this study was (i) to determine the nutritive value of oat grass supplemented with P and N plus P fertilizers and (ii) to determine the *in situ* digestion kinetics of oat grass supplemented with P and N plus P fertilizers in Nili Ravi buffalo bulls.

MATERIALS AND METHODS

The seed of oat was purchased from local market and sown in the fields of livestock experimental station Rakh Dera Chahl, Lahore. The standard agronomic practices were followed during sowing of the crop. The fodder was supplemented with P and N fertilizers designated as C; Control (without Phosphorous fertilizer) while N10P10, N15P15, N20P20, N25P25 and N30P30 represents 10 plus 10, 15 plus 15, 20 plus 20, 25 plus 25 and 30 plus 30 kg N plus P per acre, respectively. Samples of fodder harvested at 100 days of age dried at 60 °C and ground to 2mm size were analyzed for DM, CP, neutral detergent fiber (NDF), acid detergent fiber (ADF) and hemicellulose by proximate analysis (AOAC, 1990). Residual materials recovered in nylon bags after *in situ* experiment were also dried first at 60°C. Then their DM, CP, NDF, ADF and hemicellulose were determined (Van Soest *et al*., 1991).

In situ *experiments*

All the samples of oat grass supplemented with P and P plus N were oven dried at 60 °C and ground at 2 mm sieve. Approximately 10 g of each sample was weighed in nylon bag (10×23 cm) with an average pore size of 40-60 µm. The bags were closed and tied with nylon finishing line. Four cannulated Nili Ravi buffalo bulls were housed on a concrete floor in separate pen. Fresh and clean water was made available round the clock. The bulls were given 10 days adaptation period followed by four days of incubation period for *in situ* nylon bags. The bulls were fed the same diet as was being incubated in their rumen to avoid the effects of diet on rumen fermentation pattern (Faraz *et al*., 2020). The nylon bags were soaked in distilled water (39°C) for 15 min just before placing them into the rumen. The bags were then exposed to ruminal fermentation for 6, 12, 24, 48, 72 and 96 h (Sarwar and Nisa, 1999). The sample bags were placed in rumen in reverse sequence and all bags were removed at the same time to reduce variation associated with washing procedure (Faraz *et al*., 2020). These samples were washed in running tap water until water runs clear and then dried in hot air oven at 60°C. After equilibration, the bags weighed back and residue was transferred to jars and used for later analysis of crude protein, DM and NDF.

Extent of digestion, lag time and digestion rate were determined for DM, CP, NDF, ADF and hemi cellulose by subtracting residue from the amount in the bag at each time point and then regressing the natural log (Ln) of that value against time (Sarwar *et al*., 1998).

Statistical analysis

The experiment data thus obtained were subjected to statistical analysis using completely randomized design (CRD). The analysis of variance was performed to find the mean difference. Duncan's Multiple Range test was applied to estimate extent of significance ($p<0.05$) across means (Steel et al., 1997).

RESULTS

Chemical composition of oat fodder with different levels of P fertilizer

DM content of oat fodder increased ($P<0.05$) when it was supplemented with different levels of P fertilizer than that of control. However, DM content of fodder was similar in oat supplemented with different levels (P10, P15, P20, P25 and P30) of P fertilizer. CP of oat fodder was higher

(P<0.05) when it was supplemented with P30 followed by that supplemented with P20, P25, P15 and P10 (Table I).

The NDF was lower (P<0.05) in oat fodder supplemented with P30 while it was highest in control. The NDF of fodder supplemented with P10, P20 and P25 was decreased (P<0.05) in response to fertilization. Hemicelluloses of oat fodder also followed a similar trend as observed in NDF. However, ADF of fodder was similar across all treatments and did not show any effect of P supplementation (Table I).

Chemical composition of oat supplemented with NP fertilization

The DM was higher (P<0.05) in oat fodder supplemented with higher levels of NP fertilizer (N15P15, N20P20, N25P25 and N30P30) as compared to control. However, DM was similar in oat fodder supplemented with N15P15, N20P20, N25P25 and N30P30 and N10P10 and N15P15 respectively (Table II).

The CP was higher (P<0.05) in oat fodder supplemented with higher levels of NP (N30P30, N25P25 and N20P20) followed by that supplemented with N15P15 and N10P10. However, the CP was lower (P<0.05) in control. The difference among treatments N30P30, N25P25 and N20P20 and N15P15 was non significant statistically (Table II). The NDF was lower (P<0.05) in oat fodder supplemented with N30P30 while it was the highest in control. However, NDF of oat fodder supplemented with N25P25, N20P20 and N15P15 was similar and did not show any effect of NP supplementation. Hemicellulose of oat fodder showed a similar trend as was observed in NDF. The ADF of oat fodder was lower (P<0.05) in oat fodder supplemented with higher levels of NP (N30P30, N25P25 and N20P20) followed by that supplemented with medium level of NP (N15P15 and N10P10). However, the results of ADF were identical across treatments N30P30, N25P25 and N20P20 and N10P10 and N15P15. The ADF was highest (P<0.05) in control (Table II).

Effect of different levels of P fertilizer on in situ *digestion kinetics of oat fodder*

Ruminal DM and CP degradabilities were higher (P<0.05) in oat fodder supplemented with P20 and P25, followed by that supplemented with P30, P15 and P10, respectively. The lowest DM and CP degradabilities were observed in oat fodder sown without supplementation of P fertilizer. However, DM and CP degradabilities of oat fodder were similar among treatments P20 and P25 and P30, P15 and P10, respectively (Table III).

The NDF degradability at 48 h of incubation did not show any effect of P supplementation. Ruminal degradation of NDF was highest (P<0.05) in oat fodder supplemented with P30, P20 and P15 followed by that supplemented with P25 and P15, respectively. The lowest (P<0.05) ADF degradation was observed in oat fodder sown without

Table I. Composition of oat fodder with and without supplementation of phosphorous fertilizer.

Items	Control	Treatments				
		P10	P15	P20	P25	P30
DM	22.36^{b}±0.15	22.89^{a}±0.31	22.91^{a}±0.28	23.18^{a}±0.47	23.11^{a}±0.70	23.05^{a}±0.39
CP	6.56^{d}±0.13	7.00^{cd}±0.06	7.21^{c}±0.08	7.65^{b}±0.29	7.66^{b}±0.25	7.87^{a}±0.14
NDF	48.00^{a}±0.72	46.00^{b}±0.69	45.00^{bc}±1.83	44.00^{bc}±0.64	42.00^{c}±0.64	41.00^{d}±0.85
ADF	24.00±0.33	24.70±1.37	24.90±1.13	24.00±1.28	25.30±0.55	24.60±0.62
Hemicellulose	22.70^{a}±0.63	21.00^{b}±0.50	22.00^{ab}±0.24	19.40^{c}±0.66	17.30^{d}±0.50	16.10^{d}±0.43

C, Control (without phosphorous (P) fertilizer), DM, dry matter; CP, crude protein; NDF, neutral detergent fibre; ADF, acid detergent fibre. P10, P15, P20, P25, P30 represent 10, 15, 20, 25 and 30 kg P per acre, respectively. Means sharing similar letters in a row are statistically non-significant (p<0.05).

Table II. Composition of oat fodder with and without supplementation of NP fertilizers.

Items	Control	Treatments				
		N10P10	N15P15	N20P20	N25P25	N30P30
DM	22.36^{c}±0.62	24.60^{b}±0.31	25.30^{ab}±0.39	26.10^{ab}±0.54	26.70^{a}±0.50	26.90^{a}±1.01
CP	6.56^{d}±0.38	8.65^{c}±0.14	9.10^{b}±0.40	9.70^{ab}±0.19	10.06^{a}±0.25	10.08^{a}±0.16
NDF	45.00^{a}±0.57	40.00^{b}±0.53	37.00^{c}±0.35	36.00^{c}±0.65	36.00^{c}±0.36	33.00^{d}±0.58
ADF	24.00^{a}±0.69	22.00^{b}±0.27	22.70^{ab}±0.38	21.00^{c}±0.74	20.80^{c}±0.30	20.80^{c}±0.79
Hemicellulose	21.00^{a}±0.61	18.00^{b}±0.36	14.30^{c}±0.36	15.00^{c}±0.11	15.20^{c}±0.18	12.20^{d}±0.32

Means sharing similar letters in a row are statistically non-significant (p<0.05). C, control (without NP fertilizers), while N10P10, N15P15, N20P20, N25P25, N30P30 represent 10+10, 15+15, 20+20, 25+25 and 30+30 kg N+P per acre.

Table III. Effect of different levels of P fertilizer on *in situ* digestibility of oat fodder at 48 h in Nili Ravi buffalo bulls.

Items	Control	Treatments				
		P10	P15	P20	P25	P30
DM (%)	60.27^{c}±0.59	62.94^{bc}±0.90	64.99^{b}±0.91	68.33^{a}±1.74	68.08^{a}±0.90	65.37^{b}±0.54
CP (%)	58.40^{c}±0.86	59.08^{b}±0.57	59.98^{b}±1.19	64.94^{a}±0.90	63.35^{a}±1.20	62.41^{ab}±0.59
NDF (%)	59.93±1.66	59.74±0.87	59.57±0.68	61.62±1.18	59.75±1.11	60.98±2.41
ADF (%)	55.09^{d}±0.66	59.68^{c}±1.87	65.13^{ab}±0.42	67.25^{a}±0.51	62.87^{b}±1.00	66.46^{a}±0.59
Hemicellulose (%)	52.07^{c}±1.41	57.88^{b}±2.08	58.55^{b}±0.76	58.55^{b}±0.40	60.08^{ab}±1.29	62.86^{a}±0.27

For statistical detail and abbreviations, see Table I.

supplementation of P fertilizer. Hemicellulose degradation of oat fodder supplemented with higher levels of P (P30 and P25) was the highest (P<0.05) followed by that supplemented with medium P (P25, P20, P15 and P10) supplementation levels. However, Hemicellulose degradation of oat fodder did not show any effect of supplementation with P30 and P25 and P25, P20, P15 and P10, respectively. The hemicellulose degradation was the lowest (P<0.05) in oat fodder sown without supplementation (Table III).

Effect of different levels of NP fertilizers on in situ *digestion kinetics of oat fodder*

Ruminal DM degradability was higher (P<0.05) in oat fodder supplemented with N20P20 compared to control and supplemented with other levels of NP fertilizers. Ruminal DM degradation was similar in oat fodder supplemented with N30P30, N25P25 and N15P15 and N10P10 and control. The ruminal degradation of CP was the highest (P<0.05) in oat fodder supplemented with N30P30 and lowest (P<0.05) in that supplemented with N15P15, N10P10 and control (Table IV). The NDF degradation was higher (P<0.05) in all NP supplemented oat fodder as compared to control. However, the NDF degradabilities did not show any effect of NP supplementation at different levels (Table IV). Ruminal degradation of ADF was the highest (P<0.05) in oat fodder supplemented with higher NP (N30P30 and N25P25) supplementation levels followed by that supplemented with medium (N20P20 and N15P15) and lower (N10P10 and control) NP supplementation levels. Hemicellulose degradation was highest (P<0.05) in oat fodder supplemented with N30P30 while it was lowest (P<0.05) in oat fodder sown without supplementation. However, the NP supplementation did not show any effect at P25N25, N20P20, N15P15 and N10P10 levels of NP fertilizer (Table IV).

Digestion kinetics of oat fodder with different levels of P fertilizer

Dry matter extent at 96 h of incubation was higher (P<0.05) in oat fodder supplemented with P25 and P20 and was lowest (P<0.05) with P10 and control (Table V). However, DM extent was similar among treatments P20 and P25, P30 and P15, P10 and control. The DM lag (h) of oat fodder sown without supplementation was higher (P<0.05) as compared to other treatments. However, DM lag was lowest (P<0.05) in oat fodder supplemented with higher levels of P (P30, P25, P20 and P15) fertilizer (Table V). Rate of DM degradation was higher (P<0.05) in P25 and P20 supplemented fodder and was lowest in control. However, DM rate of degradation did not show any effect of P fertilizer at P30, P15 and P10, respectively (Table V). The highest level of P fertilizer failed to improve quality of fodder.

Extent of CP degradation was higher (P<0.05) in oat fodder supplemented with higher levels of P (P25 and P20) and was the lowest (P<0.05) in P30, P15, P10 supplemented and control. The CP extent was analogous across treatments P20 and P25 and P30, P15, P10 and control. The CP Lag (h) of oat fodder sown without supplementation (control) and P10 and P15 was higher (P<0.05) as compared to other treatments. However, CP lag was lowest (P<0.05) in oat fodder supplemented with highest level of P (P30). The CP rate of degradation was similar as was observed in DM rate of degradation. It was higher in P30, P25, P20 and P15 supplemented fodder and was the lowest in control and P10supplemented fodder. CP rate of degradation was similar among treatments P30, P25, P20 and P10 and control (Table V).

The extent of NDF degradation was higher (P<0.05) in oat fodder supplemented with higher levels of P (P30, P25 and P20) followed by that of supplemented with P15, P10 and was the lowest in control. The NDF extent of degradation was similar across higher levels of P (P30, P25 and P20). The NDF Lag (h) showed an inverse trend as was observed in NDF extent of degradation. The NDF lag was the highest (P<0.05) in oat fodder sown without supplementation while it was the lowest in oat fodder supplemented with higher levels of P (P30, P25 and P20). The NDF lag was similar in oat supplemented with P30,

P25 and P20, respectively. The NDF rate of degradation was higher in oat fodder supplemented with P25, while P supplementation did not show any effect NDF rate of degradation when oat fodder was supplemented with P30 and P20, P10, P15 and control (Table V).

Extent of ADF degradation was higher (P<0.05) in oat fodder supplemented with higher levels of P (P30, P25, P20, P15 and P10) and was the lowest in control. The ADF extent was similar across all levels of P supplementations. The ADF Lag (h) showed an inverse trend as was observed in ADF extent of degradation. The ADF lag was the highest in oat fodder sown without supplementation while it was the lowest in oat fodder supplemented with higher levels of P (P30, P25, P20 and P15). The ADF lag was similar across oat fodder supplemented with P30, P25, P20 and P15. The ADF rate of degradation was higher (P<0.05) in oat fodder supplemented with P25, while P supplementation did not show any effect at P30, P20 and P15 and P10 and control levels. However, ADF rate of degradation was significantly lower in oat fodder supplemented with P10 and control (Table V).

Hemicellulose extent of degradation was higher (P<0.05) in oat fodder supplemented with higher levels of P (P30, P25 and P20) and was the lowest in P10 and control. The Hemicellulose Lag (h) illustrated an inverse trend as was observed in its extent of degradation.

Table IV. Effect of different levels of NP fertilizer on *in situ* digestibility of oat fodder at 48 h in Nili Ravi buffalo bulls.

Items	Control	Treatments				
		N10P10	N15P15	N20P20	N25P25	N30P30
DM	60.27^{c}±0.59	61.65^{c}±0.96	65.20^{b}±0.52	67.90^{a}±1.37	65.75^{b}±0.40	66.30^{b}±0.58
CP	64.94^{c} ±0.90	64.30^{c} ±0.58	64.34^{c} ±0.29	66.34^{b} ±1.54	67.96^{a} ±1.73	68.28^{a} ±1.44
NDF	59.93^{b}±1.66	60.66^{a}±0.88	59.42^{a}±0.44	60.37^{a}±0.44	61.13^{a}±1.11	62.32^{a}±1.15
ADF	59.68^{c}±1.87	61.29^{c}±1.12	68.56^{b}±1.13	69.29^{b}±0.58	73.51^{a}±0.78	71.04^{ab}±0.41
Hemicellulose	51.88^{d} ±0.44	54.50^{c}±1.01	55.25^{bc}±0.75	57.94^{b}±0.22	57.88^{b}±2.08	61.57^{a}±0.45

For statistical detail and abbreviations, see Tables I and II.

Table V. Effect of different levels of P fertilizer on *in situ* digestion kinetics oat fodder in Nili Ravi buffalo bulls.

Items	Control	Treatments				
		P10	P15	P20	P25	P30
DM						
Extent, %	69.60^{c}±0.27	69.57^{c}±0.32	73.77^{b}±0.29	75.23^{a}±0.38	74.33^{a}±0.61	73.28^{b}±0.68
Lag, h	5.15^{a} ±0.59	4.48^{b} ±0.59	3.94^{bc}±0.30	3.51^{c} ±0.46	4.13^{bc} ±0.21	3.18^{c} ±0.29
Rate, %/h	3.14^{c} ±0.33	4.22^{b} ±0.16	4.15^{b} ±0.25	4.63^{a} ±0.55	4.81^{a} ±0.61	4.05^{b} ±0.05
CP						
Extent, %	68.15^{c}±0.46	69.82^{bc}±0.57	70.74^{b}±0.57	73.61^{a}±0.53	74.53^{a}±0.30	71.35^{b}±1.02
Lag, h	5.88^{a} ±1.50	5.35^{ab} ±0.63	5.25^{ab}±0.90	4.64^{b} ±0.40	3.03^{c}±1.11	2.30^{d} ±0.45
Rate, %/h	3.93^{b}±0.19	3.71^{b}±0.26	4.37^{ab}±0.33	5.17^{a}±0.37	5.12^{a}±0.47	4.40^{ab}±0.30
NDF						
Extent, %	68.72^{d}±0.34	70.49^{c}±0.36	73.27^{b}±0.64	76.67^{a}±0.88	77.10^{a}±0.34	76.41^{a}±0.23
Lag, h	6.45^{a} ±0.21	5.23^{b} ±0.94	4.63^{c} ±0.65	2.53^{d} ±0.60	2.61^{d} ±0.39	2.43^{d} ±0.54
Rate, %/h	3.49^{d}±0.32	3.64^{cd}±0.33	3.99^{c}±0.40	4.59^{b}±0.28	5.20^{a}±0.35	4.79^{b}±0.10
ADF						
Extent, %	66.89^{b}±0.28	69.08^{a}±0.41	69.86^{a}±1.08	68.90^{a}±0.36	68.09^{ab}±0.13	69.09^{a}±0.46
Lag, h	5.63^{a} ±1.30	4.06^{b} ±2.41	3.34^{c} ±0.06	3.34^{c} ±0.06	2.71^{cd} ±0.63	2.29^{d} ±0.41
Rate, %/h	3.34^{c}±0.10	3.42^{c}±0.25	4.06^{b}±0.45	4.26^{b}±0.13	5.60^{a}±0.60	3.94^{b}±0.24
Hemicellulose						
Extent, %	65.90^{c}±0.32	66.44^{c}±0.60	68.87^{b}±0.64	71.60^{a}±0.83	72.20^{a}±0.86	71.69^{a}±0.76
Lag, h	5.57^{a} ±0.51	4.48^{b} ±0.29	4.62^{b} ±0.98	4.47^{b} ±0.26	3.70^{c} ±0.34	2.51^{d} ±0.52
Rate, %/h	3.32^{c}±0.18	3.46^{c}±0.35	3.90^{b}±0.17	4.89^{a}±0.57	4.93^{a}±0.40	4.01^{b}±0.21

For statistical detail and abbreviations, see Tables I and II.

Hemicellulose rate of degradation was higher (P<0.05) in oat fodder supplemented with P25 and P20, while P supplementation did not show any effect at P30 and P15, P10 and control levels. However, rate of hemicellulose degradation was lower (P<0.05) in oat fodder supplemented with P10 and control (Table V).

Digestion kinetics of oat fodder supplemented with different levels of nitrogen and phosphorus fertilizer

The extent of DM degradation at 96h of incubation was higher (P<0.05) in oat fodder supplemented with N25P25 and N20P20 fertilizers and was the lowest (P<0.05) in N10P10 supplemented oat and control. However, extent of DM degradation was similar among treatments N20P20 and N25P25, N30P30 and N15P15 and N10P10 and control, respectively. The DM Lag (h) of oat fodder sown without supplementation and N10P10 was higher (P<0.05) as compared to other treatments. However, DM lag was the lowest (P<0.05) in oat fodder supplemented with higher levels of NP (N30P30, N25P25 and N20P20) fertilizers. Rate of DM degradation was higher in N20P20 supplemented oat fodder and was the lowest significantly in control. However, DM rate of degradation was similar across treatments N30P30, N25P25 and N15P15. It was observed that the highest level of phosphorus and Nitrogen failed to improve quality of the oat fodder (Table VI).

Extent of CP degradation was higher (P<0.05) in oat fodder supplemented with higher levels of NP (N30P30 and N25P25) fertilizers followed by that of supplemented with N20P20 and N15P15 and was the lowest (P<0.05) in N10P10 supplemented and control. The CP extent was analogous across treatments N30P30 and N25P25, N20P20 and N15P15 and N10P10 and control. The CP Lag (h) of oat fodder sown without supplementation was higher (P<0.05) as compared to other treatments while it was the lowest (P<0.05) in oat fodder supplemented with N25P25. However, CP lag was similar in treatment N30P30, N20P20 and N15P15, respectively. The CP rate of degradation was higher in oat fodder supplemented with N25P25 and was the lowest in control. It was similar among treatments P30N30 and P20N20 and P15N15 and P10N10 supplemented fodder (Table VI).

Extent of NDF degradation was higher (P<0.05) in oat fodder supplemented with all levels of NP fertilizers and was the lowest in control. The NDF extent was alike across all levels of NP supplementation. The NDF Lag (h) showed an inverse trend as was observed in NDF extent of degradation. The NDF rate of degradation was higher (P<0.05) in oat fodder supplemented with N25P25, while it was similar in oat fodder supplemented with N30P30 and N20P20 and N15P15, N10P10 and control (Table VI).

Table VI. Effect of different levels of NP fertilizer on *in situ* kinetics of oat fodder in Nili Ravi buffalo bulls.

Nutrients	Control	Treatments				
		N10P10	N15P15	N20P20	N25P25	N30P30
DM						
Extent, %	69.60^{c}±0.27	69.47^{c}±0.58	70.70^{bc}±0.66	74.24^{a}±0.77	74.34^{a}±0.87	71.73^{b}±0.51
Lag, h	5.33^{a}±0.58	5.15^{ab}±0.59	4.59^{b}±0.31	3.86^{bc}±0.34	4.08^{bc}±0.12	3.14^{c}±0.30
Rate, %/h	3.45^{d} ±0.57	3.91^{c} ±0.37	4.53^{b} ±0.22	4.81^{a} ±0.61	4.52^{b} ±0.26	4.58^{b} ±0.08
CP						
Extent, %	68.15^{c}±0.46	72.77^{c}±1.62	73.78^{b}±0.69	74.20^{b}±0.87	78.36^{a}±2.08	77.77^{a}±2.76
Lag, h	4.01^{a} ±0.18	3.50^{b}±0.62	2.72^{c}±1.24	2.70^{c}±1.13	2.09^{d}±0.92	2.30^{cd}±0.45
Rate, %/h	3.38^{d} ±0.39	3.71^{c}±0.27	3.78^{c}±0.19	4.73^{b}±0.22	5.12^{a}±0.47	4.62^{b}±0.45
NDF						
Extent, %	68.72^{c}±0.34	73.55^{b}±0.66	76.83^{ab}±1.10	77.76^{a}±1.77	78.80^{a}±0.79	80.12^{a}±1.85
Lag, h	4.82^{a} ±0.44	3.63^{b} ±0.33	3.51^{b} ±0.31	3.17^{c} ±0.54	2.42^{d} ±0.06	2.61^{d} ±0.39
Rate, %/h	3.05^{c} ±0.34	3.06^{c} ±0.03	3.18^{c} ±0.11	4.70^{b} ±0.45	5.20^{a} ±0.35	4.57^{b} ±0.23
ADF						
Extent, %	69.08^{b} ±0.41	72.73^{a}±0.97	72.74^{a}±1.23	72.38^{a}±0.72	73.55^{a}±1.41	73.23^{a}±0.82
Lag, h	3.34^{c}±0.06	4.56^{b}±0.19	6.11^{a}±0.39	3.69^{c}±0.13	1.69^{d}±0.40	1.22^{d}±0.53
Rate, %/h	2.67^{c} ±0.13	2.71^{c} ±0.12	3.06^{d} ±0.00	4.54^{b} ±0.15	5.60^{a} ±0.60	4.10^{c} ±0.01
Hemicellulose						
Extent, %	67.67^{c}±0.46	68.87^{bc}±0.64	69.40^{b}±0.59	71.69^{ab}±0.50	71.26^{ab}±1.52	72.66^{a}±0.40
Lag, h	3.91^{a}±0.23	3.77^{a}±0.19	3.54^{b}±0.18	3.47^{b}±0.13	2.31^{c}±0.48	2.51^{c}±0.52
Rate, %/h	2.80^{d}±0.17	2.77^{d}±0.23	3.63^{c}±0.25	4.55^{b}±0.23	4.93^{a}±0.40	4.24^{b}±0.08

For statistical detail and abbreviations, see Tables I and II.

Extent of ADF degradation was higher (P<0.05) in oat fodder supplemented with all the levels of NP and was the lowest in control. However, ADF extent of degradation was alike across all levels of NP supplementations. The ADF lag was the highest in oat fodder supplementation with N10P10 while it was the lowest in that supplemented with higher levels of NP (N30P30 and N25P25) fertilizers. The ADF lag was similar across oat fodder supplemented with N30P30 and N25P25, N20P20 and control. The ADF rate of degradation was higher (P<0.05) in oat fodder supplemented with N25P25 followed by that of supplemented with N20P20, N30P30 and N15P15, respectively while it was the lowest in N10P10 supplemented fodder and control (Table VI).

The extent of hemicellulose degradation was higher (P<0.05) in oat fodder supplemented with all levels of NP supplementation as compared to control. Hemicellulose Lag (h) illustrated an inverse trend as was observed in extent of degradation. Hemicellulose rate of degradation was higher (P<0.05) in oat fodder supplemented with N25P25 fertilizer, while it was similar in oat fodder supplemented with N30P30 and N20P20, N10P10 and control, respectively. However, hemicellulose rate of degradation was lowest (P<0.05) in oat fodder supplemented with N10P10 and control (Table VI).

DISCUSSION

chemical composition of oat fodder

The results of current study revealed that DM content of oat fodder was increased in response to P fertilization. These results were consistent to the findings of Rashid *et al*. (2007) who also reported that P fertilization resulted in increased DM yield of Sorghum fodder. Similar results have also been reported by Pant *et al*. (2004) that forage yield was increased in response to P fertilization. They also reported that using P 10 kg/hac/year improved forage production. Desai and Deore (1980) also reported higher DM yields with P fertilization of cowpea fodder as compared to sown without P fertilization. The increased fodder production might be due to the optimum supply of P to the developing fodder seedlings during early growth stage of crop. As the tropical soils are deficient in P and the additional P supply through fertilizer stored in the soil and this stored P becomes available slowly and might be used to meet P requirements of developing seedlings of fodder (Saarela *et al*., 2003).

The nutritive value of oat fodder was increased with NP fertilization. The findings of current study were consistent with results as reported by Desai and Deore (1980) that application of NP fertilizers (90 kg N/ ha and 40 kg P/ha) resulted in higher DM and CP production than application of N or P fertilizer alone. Similarly, Sultan *et al*. (2008) observed that the use of NP fertilizer not only increased the biomass yield of grasses (Dichanthium Annulatum and Cenchrus Sigerus Vahl) but also improved their nutritive value. They further justified that supplementation of NP fertilizers results in improvement of NP status of the soil and also enhanced the yield of fodder (Khan *et al*., 2000) by providing essential nutrients for plant growth. Moreover, application of NP fertilizer favors transformation of carbohydrates into protein (Khan *et al*., 2000) thus improving the nutritive value of the plant.

In situ *digestibility*

Results of the current study have supported the findings of the Puoli *et al*. (1992) who reported that DM and NDF digestibilities were higher in switch grass fertilized with N plus Sulfur fertilizers. This might be due to positive impact of N fertilization on growth of the grass which improved its DMD due to decrease in ruminal retention time. Contrary to these, Johnson *et al*. (2001) did not find any effect of N fertilization (0, 39, 78, 118 and 157 kg N/hac) on *in vitro* organic matter digestibility of Bahia grass whereas digestibility of star grass was increased linearly and Bermuda grass quadratically with increasing levels of N fertilization. Similarly, Hughes and Haslemore (1981) reported that application of N fertilizer had non-significant effect on CP digestibility in oat forage. Whereas, Nichols (1990) observed that N fertilization (135 kg N/ hac) decreased 5.2% digestibility of meadow vegetation compared to those without N fertilization. This decrease in digestibility was due to formation of stein tissues which constitute 70% of the crop weight and characterized with lower digestibility.

In situ *digestion kinetics*

The supplementation of oat fodder with N and P fertilizers had positive impact on its nutritive value and digestibility. But the highest levels of phosphorus failed to improve quality of the fodder. Ruminal DM, CP, NDF, ADF and hemicellulose lag time was lower in bulls fed fodder sown with application of P or PN fertilizers compared to those sown without supplementation. The probable reason might be that the supplementation of P or PN fertilizers improved nutritive value of the fodder by improving DM and CP which enhanced its utilization by the animals and hence the digestibility was higher. Lima *et al*. (1999) also reported that N fertilization affected the N contents of the plant and increased degradation rate of NDF due to increased CP contents of timothy grass (Nordheim and Volden, 2009).

Increasing levels of N fertilization also resulted in higher degradation rate of cell walls (Bélanger and

McQueen, 2003). The results of current study also supported the findings of Puoli *et al*. (1992) who reported that N fertilization reduced DM and NDF ruminal turnover times. Similarly, Romero *et al*. (1976) found that supplementing N increased digesta rate of passage out of the rumen, increased dietary N intake and stimulates rumen microbial growth (Preston and Leng, 1987; Poppi and McLennan, 1995) resulting in increased rate of disappearance, extent of digestion and lower lag time. In contrast, Messman *et al*. (1991) found that N fertilization had no effect on extent of fiber digestion. Cell wall contents and their digestibility remained unaffected with N fertilization (Deinum, 1984). In conclusion, supplementation of 20-25kg/acre phosphorus or phosphorus plus nitrogen fertilizers in oat fodder not only improved nutritive value but also increased nutrient digestibility, rate of disappearance and extent of digestion of oat fodder in Nili Ravi buffalo bulls.

CONCLUSION

Supplementation of oat fodder with 20–25 kg/acre phosphorous or phosphorous plus nitrogen not only improved the nutritive value but also increased nutrient digestibility, rate of disappearance and extent of digestion of oat fodder in Nili Ravi buffalo bulls.

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Statement of conflict of interest

The authors have declared no conflict of interest.

REFERENCES

AOAC, 1990. *Official methods of analysis*. Assoc. Anlyt. Chemists, 15th Ed. Arlington Virginia, U.S.A.

Bélanger, G., McQueen, R.E., 2003. Analysis of the nutritive value timothy grown with varying N nutrition. *Grass Forage Sci*., **53**: 10 –119. https://doi.org/10.1046/j.1365-2494.1998.5320109.x

Bulla, R.J., Lechtenberg, V.L., and Holt, D.A., 1997. *Potential of the world's forages for ruminant animal production*. Winrock International Livestock Research and Training Center pit Jean Mountain, Morrilton, Arkansas, USA.

Deinum, B., 1984. *Chemical composition and nutritive value of herbage in relation to climate*. Proc. 10th Gen. Meet Eur. Grassl Fed, Assoc., Norway. pp. 338-350.

Desai, S.N. and Deore, D.D., 1980. Performance of forage sorghum varieties (*Sorghum bicolor*) under nitrogen fertilizer. *Forage Res*., **6**: 35-38.

Dost, M., 1997. *End of assignment report on fodder component*. UNDP/FAO project PAK/86/027. FAO/UNDP, Gilgit, Pakistan.

Faraz, A., Tauqir, N.A., Bilal, R.M., Ahmad, F. and Waheed, A., 2020. In situ nutritional evaluation of maize varieties harvested at different growth stages. *Adv. Anim. Vet. Sci*., **8**: 455-466. https://doi.org/10.17582/journal.aavs/2021/9.3.330.334

Fisher, D.S., Burns, J.C. and Pond, K.R., 1989. Kinetics of cell wall disappearance and in vivo digestion. *Agro. J*., **81**: 25-33. https://doi.org/10.2134/agronj1989.00021962008100010005x

Grant, C.A., Flaten, D.N., Tomasiewicz, D.J., and Sheppard, S.C., 2001. The importance of early Seasons phosphorus nutrition. *Can. J. Pl. Sci*., **81**: 211–224. https://doi.org/10.4141/P00-093

Hughes, K.A. and Haslemore, R.M., 1981. Winter oats changes in nutritive value during development of the crop. *Proc. Agron. Soc. N. Z*., **11**: 41-44.

Johnson, L., Harrison, J.H., Davidson, D., Mahanna, W.C., Shinners, K., and Linder, D., 2001. Corn silage management: effect of maturity, inoculation, and mechanical processing on pack density and aerobic stability. *J. Dairy Sci*., **85**: 434–444. https://doi.org/10.3168/jds.S0022-0302(02)74092-7

Khan, H., Khan, M., Rasul, K., Majeed, A. and Safi, F.A., 2000. Effect of different levels of nitrogen alone and in combination with constant doses of phosphorus and potassium on growth and yield of okra (*Hibiscus esculentus* L.) Cv. T-13 under agro-climatic conditions of Mingora, Swat. *Pak. J. biol. Sci*., **3**: 2101-2104. https://doi.org/10.3923/pjbs.2000.2101.2104

Lima, G.F.C., Sollenberger, L.E., Kunkle, W.E., Moore, J.E. and Hammond, A.C., 1999. Nitrogen fertilization and supplementation effects on performance of beef heifers grazing limpo grass. *Crop Sci*., **39**: 1853–1858. https://doi.org/10.2135/cropsci1999.3961853x

Mahmud, K., Ahmad, I. and Ayub, M., 2003. Effect of nitrogen and phosphorus on the fodder yield and quality of two sorghum cultivars (*Sorghum bicolor* L.). *Int. J. Agric. Biol*., 1560-8530.

Messman, M.A., Weiss, W.P. and Erickson, D.O., 1991. Effect of nitrogen fertilization and maturity of bromegrass on *in situ* ruminal digestion kinetics of fiber. *J. Anim. Sci*., **69**: 1151. https://doi.org/10.2527/1991.6931151x

Nichols, J.T., Reece, P.E., Hergert, G.W. and Moser, L.E., 1990. Yield and quality response of sub irrigated meadow vegetation to nitrogen, phosphorus and sulfur fertilization. *Agron. J.*, **54**: 47–52. https://doi.org/10.2134/agronj1990.00021962008200010010x

Nordheim, H. and Volden, H., 2009. Effect of maturity stage, nitrogen fertilization and seasonal variation on ruminal degradation characteristics of neutral detergent fibre in timothy (*Phleum pratense* L.). *Anim. Feed Sci. Technol.,* **149**: 30-59.

Pant, H.K., Mislevy, P. and Rechcgi, J.E., 2004. Effects of phosphorus and potassium on forage nutritive value and quantity environmental implications. *Agron. J.*, **96**: 1299–1305. https://doi.org/10.2134/agronj2004.1299

Poppi, D.P. and McLennan, S.R., 1995. Protein and energy utilization by ruminants at pastures. *J. Anim. Sci.*, **73**: 278-290. https://doi.org/10.2527/1995.731278x

Preston, T.R. and Leng, R.A., 1987. *Matching ruminant production systems with available resources in the tropics and subtropics.* Penambul Books Ltd: Armidale NSW, Australia.

Puoli, J.R., Reid, R.L. and Belesky, D.P., 1992. Photosensitization in lambs grazing switchgrass. *Agron. J.*, **84**: 1077-1080. https://doi.org/10.2134/agronj1992.00021962008400060033x

Rashid, M., Ranjha, A.M. and Rahim, A., 2007. Model based P fertilization to improve yield and quality of sorghum (*Sorghum bicolor* L.) fodder on an ustochrept soil. *Pak. J. agric. Sci.*, **44**: 221-227.

Romero, A.V., Siebert, B.D., and Murray, R.M., 1976. A study on the effect of frequency of urea ingestion on the utilization of low-quality roughages by steers. *Aust. J. exp. Agric.*, **16**: 308–314. https://doi.org/10.1071/EA9760308

Rotz, C.A., Sharpley, A.N., Gburek, W.J., Satter, L.D., and Sanderson, M.A., 2002. Production and feeding strategies for phosphorus management on dairy farms. *J. Dairy Sci.*, **85**: 3142–3153. https://doi.org/10.3168/jds.S0022-0302(02)74402-0

Saarela, I., ärvi, A., Hakkola, H. and Rinne, K., 2003. Phosphorus status of diverse soils in Finland as influenced by long-term P fertilization I. Native and previously applied P at 24 experimental sites. *Agric. Fd. Sci. Finland*, **12**: 117-132. https://doi.org/10.23986/afsci.5747

Sarwar, M. and Nisa, M.U., 1999. Effect of nitrogen fertilization and stage of maturity of Mott grass (*Penisetum perpureum*) on its chemical composition, dry matter intake, ruminal characteristics and digestibility in buffalo bulls. *Asian-Aust. J. Anim. Sci.*, **12**: 1040-1041. https://doi.org/10.5713/ajas.1999.1035

Sarwar, M., Nisa, M. and Sial, M.A. 1998. Factors affecting digestion kinetics of forages in ruminants (A review). *Pak. Vet. J.*, **18**: 58-62.

Sarwar, M., Khan, M.A., and Iqbal, Z., 2002. Feed resources for livestock in Pakistan. *Int. J. Agric. Biol.*, **4**: 245-257.

Steel, R.G.D., Torrie, J.H. and Dicky, D.A., 1997. *Principles and procedures of statistics*. A biometric approach 3rd Ed. McGraw Hill Book Co., New York, USA.

Sultan, J.I., Inam-Ur-Rahim, Nawaz, H., Yaqoob, M. and Javed, I., 2008. Nutritional evaluation of fodder tree leaves of northern grasslands of Pakistan. *Pak. J. Bot.*, **40**: 2503-2512.

Van Soest, P.J., Robertson, J.B. and Lewis, B.A., 1991. Methods for dietary fiber, neutral detergent fiber, and non-starch polysaccharides in relation to animal nutrition. *J. Dairy Sci.*, **74**: 3583-3597. https://doi.org/10.3168/jds.S0022-0302(91)78551-2