Research Paper

Effects of seeding rate, fertilizing time and fertilizer type on yield, nutritive value and silage quality of whole-crop wheat

Efectos de tasa de siembra, momento de aplicación y tipo de fertilizante en el rendimiento, el valor nutritivo y la calidad del ensilaje de trigo integral

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Abstract

Whole-crop wheat (WCW) is rich in nutrients and is widely used as a forage crop. This study consisted of 2 experiments: Experiment 1 studied the yield, nutritive value and silage quality of WCW at 3 seeding rates (320 kg/ha, S₃₂₀; 385 kg/ha, S₃₈₅; and 450 kg/ha, S₄₅₀) and different fertilizing times, i.e. 60% at seedling stage and the remaining 40% at the jointing stage vs. heading stage; and Experiment 2 examined the yield, nutritive value and silage quality of WCW receiving different fertilizer types, i.e. urea, compound fertilizer (N:P:K) and urea + compound fertilizer (all iso-nitrogenous). With the increased seeding rate, dry matter (DM) and crude protein (CP) yields tended to increase, but relative feed value tended to decrease. Experiment 1: there was no significant interaction between time of applying the second fertilizer dose and seeding rate in terms of concentrations of CP, crude fiber, ether extract, crude ash, nitrogen-free extract, neutral detergent fiber (NDF) and acid detergent fiber (ADF) in wheat (P>0.05). However, a significant interaction between fertilizing time and seeding rate was observed in terms of silage fermentation quality (pH, lactic acid, butyric acid and NH₃-N concentrations) (P<0.05). Experiment 2: DM yield, CP yield and concentrations of CP, ADF and water-soluble carbohydrate were not affected by fertilizer type (P>0.05). Fertilizer type had significant effects on pH of silage and concentrations of organic acids (except propionic acid) and NH₃-N in WCW silage (P<0.05). Under the present study conditions, considering DM yield, nutrient composition and silage fermentation quality, an optimal seeding rate of wheat for forage appears to be about 385 kg/ha. N fertilizer should be applied at the seedling stage and jointing stage. Although applying a mixture of urea and compound fertilizer had no significant effects on yield and nutritive value of WCW relative to applying urea alone, it did improve silage fermentation quality. Results may differ on different soils.

Keywords: Fertilizer application, nutritional composition, seeding rate, whole-crop wheat, yield.

Resumen

El trigo integral (WCW) es rico en nutrientes y se usa ampliamente como cultivo forrajero. Este estudio consistió en 2 experimentos: el Experimento 1 estudió el rendimiento, valor nutritivo y calidad del ensilaje de WCW a 3 tasas de siembra (320 kg/ha, S₃₂₀; 385 kg/ha, S₃₈₅; y 450 kg/ha, S₄₅₀) y diferentes tiempos de fertilización: el 60% en la etapa de plántula y el 40% restante entre las etapas de unión y encabezado. El Experimento 2 examinó el rendimiento, el valor nutritivo y la calidad del ensilaje de WCW que recibieron diferentes tipos de fertilizantes: urea, fertilizante compuesto (N:P:K) y urea + fertilizante compuesto (todos iso-nitrogenados). Con el aumento de la tasa de siembra, los rendimientos de materia seca (DM) y proteína cruda (CP) tendieron a aumentar, pero el valor relativo del alimento tendió a disminuir. Experimento 1: no hubo interacción significativa entre el tiempo de aplicación de la segunda dosis de fertilizante y la tasa de siembra en términos de concentraciones de CP, fibra bruta, extracto de éter, ceniza bruta, extracto libre de nitrógeno, fibra detergente neutra (NDF) y fibra detergente ácida (ADF) en trigo (P>0.05). Sin embargo, se observó una

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significativa interacción entre el tiempo de fertilización y la tasa de siembra en términos de la calidad de fermentación del ensilaje (pH, ácido láctico, ácido butírico y concentraciones de NH₃-N) (P <0.05). Experimento 2: El rendimiento de DM, el rendimiento de PC y las concentraciones de PC, ADF y carbohidratos solubles en agua no se vieron afectados por el tipo de fertilizante (P>0.05). El tipo de fertilizante tuvo efectos significativos sobre el pH del ensilaje y las concentraciones de ácidos orgánicos (excepto ácido propiónico) y NH₃-N en el ensilaje WCW (P<0.05). En las condiciones del presente estudio, considerando el rendimiento de DM, la composición de nutrientes y la calidad de la fermentación del ensilaje, una tasa óptima de siembra de trigo para forraje parece ser de unos 385 kg / ha. El fertilizante N debe aplicarse en la etapa de plántula y en la etapa de unión. Aunque la aplicación de una mezcla de urea y fertilizante compuesto no tuvo efectos significativos sobre el rendimiento y el valor nutritivo de WCW en relación con la aplicación de urea sola, sí mejoró la calidad de la fermentación del ensilado. Los resultados pueden diferir en diferentes suelos.

Palabras clave: Aplicación de fertilizantes, composición nutricional, rendimiento, tasa de siembra, trigo integral.

Introduction

The demand for animal proteins in China is rapidly growing with the improvement of living standards and the change of food consumption habits. However, the development of animal husbandry is usually restricted by shortage of herbage supply (Liu et al. 2012). Therefore, there is increasing need to develop and utilize new herbage resources or find new lands to grow herbage crops. Whole-crop wheat (Triticum aestivum) (WCW) has relatively high nutritional value (Sprague et al. 2015) and total DM intake of WCW diets exceeded that of grass diets (Günal et al. 2018). In order to improve the economic returns from farms, more and more WCW is being planted instead of grass (Huuskonen et al. 2017), often as a specialized forage wheat or as an addition within forage production systems. For example, WCW is processed into hay and silage to feed beef cattle in Finland (Huuskonen et al. 2017), while in Australia, dual-purpose wheat is often planted and used for grazing to alleviate winter feed shortages (Sprague et al. 2015). In Oklahoma, USA, in response to the lack of forage in winter, large areas are used to grow wheat for animal forage in the form of whole plants at maturity (Kim and Anderson 2015). Although wheat has been widely used as forage, few studies have focused on optimal planting techniques. Previous studies on forage wheat have concentrated on variety screening (Li 2015), nitrogen (N) application rate, harvest time (Xie 2012) and silage utilization (Filya 2003; Shaani et al. 2017).

Among cultivation measures, the factors that have the greatest impact on forage yield and nutritive value are seeding rate and N fertilizer management (Guo et al. 2017). Li (2015) found that, at a seeding rate of 260 kg/ha, there was still potential for dry matter yield (DMY) of forage wheat to increase if seeding rate was increased. However, the number of wheat spikes tended to decrease with increases in seeding rate (Yang 2011). In order to obtain data on optimal seeding rates to achieve a desirable balance between yield and quality of forage wheat, further research is needed. While Pan et al. (1999) found that, in terms of DMY, the Law of Diminishing Returns operated with increase in N application rate and yield even declined past a certain application rate, application of N increased crude protein (CP) concentration, in vitro dry matter digestibility and silage fermentation quality of forage wheat (Li et al. 2016).

Not only is amount of N applied important but also timing of the application is critical. Accumulation of DM in wheat occurs mainly during the period from jointing to maturity, accounting for 70% of the total DM yield (<u>Wu and Cui 2000</u>). Applying N fertilizer at the jointing stage increases the leaf area index of wheat, accumulates more DM during the vegetative period and increases the number of tillers (<u>Ravier et al. 2017</u>). However, little is known of the efficiency of fertilizer use when applied to wheat close to flowering (heading stage).

In winter, fields are fallowed after the harvest of late rice in Southern China, which would allow the planting of a winter-forage crop (Cinar et al. 2020). However, frequent cultivation leads to low nutrient levels in the soil, so producers often use compound fertilizer to meet the needs of winter-forage crops. The effects of seeding rate, fertilizing time and fertilizer type on yield, nutritive value and silage quality of WCW have not been explored. Therefore, in this study, we aimed to compare the effects of different seeding rates and timing of fertilizer application on yield and nutritive value of forage produced. We hypothesized that: (i) high seeding rate would increase both yield and nutritive value of forage; (ii) applying part of the fertilizer at jointing stage is better than applying all at heading stage; and (iii) applying urea with compound fertilizer would increase yield and nutritive value of WCW to higher levels than urea or compound fertilizer alone.

Materials and Methods

Experimental sites

Experiment 1 was carried out at Meitan Experimental Field of Agricultural Science Institute of Qingyuan (23°42′ N, 115°50′ E), Guangdong Province, China. The site is located within a subtropical monsoon humid climate zone with an annual average temperature of 22.3 °C. The hottest month is July with a monthly average temperature of 31.4 °C, while the coldest month is January with a monthly average temperature of 14.0 °C. The annual average rainfall and sunshine time are 1,842 mm and 2,245 hours, respectively.

Experiment 2 was carried out at Ningxi Experimental Field of South China Agricultural University (23°14′ N, 113°38′ E), Zengcheng, Guangzhou, Guangdong Province, China. This site is also located within a subtropical monsoon humid climate zone with an annual average temperature of 21.6 °C. The hottest month is July with a monthly average temperature of 29.4 °C, while the coldest month is January with a monthly average temperature of 13.3 °C. The annual average rainfall and sunshine time are 1,968 mm and 2,107 hours, respectively.

Meteorological data for the 2 sites during the study plus the medium-term mean data are presented in Table 1.

For the two experimental sites, the general cropping systems are early rice in spring (summer harvest) and late rice in summer (autumn harvest), then either fallowing or planting winter forage crops (to be harvested in spring of the following year). Soil types are cinnamon soil for Meitan Experimental Field and paddy soil for Ningxi Experimental Field (Zhang et al. 2014). Before sowing the forage wheat, 5 soil cores (each 2.5 cm diameter) were randomly excavated and mixed to give a composite sample for determining soil chemical properties. The soil chemical composition was similar at both sites (Table 2).

Wheat planting and management

Wheat phenology was regularly monitored based upon the Decimal Code (DC) (Zadoks et al. 1974). In Experiment 1, a factorial arrangement of timing of N application (jointing vs. heading) \times seeding rate was utilized. A compound fertilizer (N:P:K, 15:6:8) was applied at 150 kg/ha with 60% at the seedling stage (DC13) and 40% at the jointing (DC31)or heading stage (DC41), with 3 seeding rates, i.e. the recommended rate of 320 kg/ha (S₁₀₀) (Li 2015) and increased rates of +20% (384 kg/ha; S₁₂₀) and +40% (448 kg/ha; S_{140}) (Table 3). In Experiment 2, urea, compound fertilizer (N:P:K, 15:6:8) and a combination of urea and compound fertilizer (5:5) were compared. All treatments were designed to apply 150 kg N/ha in total. A standard seeding rate of 385 kg/ha was used. Sixty percent of the fertilizer was applied at the seedling stage (DC13) and the remaining 40% at the jointing stage (DC31).

In Experiment 1, the planting and harvesting dates of wheat were 8 November 2014 and 10 March 2015, respectively, while in Experiment 2, the planting and harvesting dates were 10 November 2014 and 25 March 2015, respectively. The wheat variety was Shimai No.1 (seed germination rate 98%, 53 mg per seed). In both experiments there were 3 replicates of the above treatments, arranged as a randomized block, and each plot was 12 m^2 (3 × 4 m).

|--|

| Parameter | Meitan EF ¹ | Ningxi EF |
|------------------------------|------------------------|-----------|
| Organic matter (g/kg) | 4.20 | 2.75 |
| Total nitrogen (g/kg) | 1.35 | 1.38 |
| Total phosphorus (g/kg) | 1.58 | 2.89 |
| Total potassium (g/kg) | 16.6 | 18.7 |
| Available nitrogen (mg/kg) | 92.4 | 84.1 |
| Available phosphorus (mg/kg) | 63.4 | 67.6 |
| Available potassium (mg/kg) | 132 | 150 |
| pH | 5.27 | 5.15 |

¹EF: Experimental field

| Location | Parameter | Nov | Dec | Jan | Feb | Mar |
|-----------------------------|---------------------------------|------|------|------|------|------|
| Meitan experimental field | Mean temperature (°C) | 19.6 | 16.7 | 16.2 | 15.6 | 18.3 |
| | Rainfall (mm) | 64 | 14 | 10 | 18 | 125 |
| | No. of days with rainfall (d) | 9 | 5 | 9 | 8 | 17 |
| Ningxi experimental field | Mean temperature (°C) | 19.9 | 16.6 | 14.1 | 14.5 | 19.9 |
| | Rainfall (mm) | 59 | 10 | 136 | 12 | 69 |
| | No. of days with rainfall (d) | 8 | 1 | 8 | 5 | 8 |
| Meitan experimental field – | Mean temperature (°C) | 21.1 | 17.3 | 13.6 | 16.0 | 19.8 |
| 20-year mean | Rainfall (mm) | 38 | 44 | 41 | 20 | 103 |
| | No. of days with rainfall (np-) | 5 | 9 | 8 | 4 | 12 |
| Ningxi experimental field – | Mean temperature (°C) | 20.4 | 15.6 | 14.4 | 15.2 | 18.7 |
| 20-year mean | Rainfall (mm) | 46 | 50 | 56 | 22 | 116 |
| - | No. of days with rainfall (no.) | 7 | 8 | 6 | 7 | 13 |

| periment Planting Fertilizing date and Decimal Code | | | | | Harvesting | date and | Crop growth time | | |
|---|--|---|---|--|---|--|--|---|--|
| date | | | | | | | Decimal | l Code | (No. of days) |
| | Seedling | Decimal | Jointing | Decimal | Heading | Decimal | Date | Decimal | |
| | stage | Code | stage | Code | stage | Code | | Code | |
| 08/11/2014 | 16/12/2014 | DC13 | 18/02/2015 | DC31 | 27/02/2015 | DC41 | 10/03/2015 | DC77 | 122 |
| | | | | | | | | | |
| | | | | | | | | | |
| 10/11/2014 | 24/12/2014 | DC13 | 22/02/2015 | DC31 | - | - | 25/03/2015 | DC87 | 135 |
| | | | | | | | | | |
| | Planting date 08/11/2014 10/11/2014 | Planting date Seedling stage 08/11/2014 16/12/2014 10/11/2014 24/12/2014 | Planting date Ferti- Seedling Decimal Code 08/11/2014 16/12/2014 DC13 10/11/2014 24/12/2014 DC13 | Planting date Seedling 08/11/2014 16/12/2014 DC13 18/02/2015 10/11/2014 24/12/2014 DC13 22/02/2015 | Planting date Seedling stage 08/11/2014 10/11/2014 24/12/2014 DC13 Decimal Code Stage Code Stage Code Stage Code Stage Code Stage Code Stage Code Stage Code Stage Code Stage Code DC13 18/02/2015 DC31 | Planting dateFertilizing date and Decimal CodedateSeedling stageDecimal CodeJointing stageDecimal CodeHeading stage08/11/201416/12/2014DC1318/02/2015DC3127/02/201510/11/201424/12/2014DC1322/02/2015DC31- | Planting dateFertilizing date and Decimal CodedateSeedling stageDecimal CodeJointing stageDecimal CodeHeading stageDecimal Code08/11/201416/12/2014DC1318/02/2015DC3127/02/2015DC4110/11/201424/12/2014DC1322/02/2015DC31 | Planting dateFertilizing date and Decimal CodeHarvesting DecimaldateSeedling stageDecimal CodeJointing stageDecimal CodeHeading StageDecimal Code08/11/201416/12/2014DC1318/02/2015DC3127/02/2015DC4110/03/201510/11/201424/12/2014DC1322/02/2015DC3125/03/2015 | Planting dateFertilizing date and Decimal CodeHarvesting date and Decimal CodedateSeedling stageDecimal CodeJointing stageDecimal CodeHeading stageDecimal CodeDateDecimal Code08/11/201416/12/2014DC1318/02/2015DC3127/02/2015DC4110/03/2015DC7710/11/201424/12/2014DC1322/02/2015DC3125/03/2015DC87 |

Table 3. Agricultural operation dates and Decimal Code of wheat development stages.

Field investigation and sampling

In Experiment 1, wheat was harvested at the milk stage (DC77), while in Experiment 2 harvesting was at the soft dough stage (DC87). Fifteen wheat plants per plot were randomly selected to determine plant height and tiller number, and the average value was calculated. In each plot a 1 m² (1 × 1 m) site was selected at random and forage was harvested at 5 cm from ground level to measure yield. All harvested material was taken back to the laboratory and cut into 2–3 cm pieces by a forage chopper. Fresh material was used to determine microorganisms present and to make silage.

Silage making

After being cut into pieces, fresh material from each plot was mixed uniformly and a 200 g sample was packed into a 30×20 cm polyethylene silage bag, air was removed and the bag was sealed with a vacuum packer (Sinbo Vacuum Sealer, Hong Tai Home Electrical Appliance Co. Ltd, Hong Kong, China) (Xie et al. 2012). Silage packs were stored in the dark at room temperature for 60 d, before being analyzed for silage fermentation quality.

Chemical and microbial analyses

Crop material was dried at 70 °C for 48 hours in an oven with forced-air circulation for determination of DM concentration. N concentration was determined by the Kjeldahl method (Nitrogen analyzer KN680, Shandong Jinan Alva Instrument Co. Ltd, Jinan, China), and ammonia nitrogen (NH₃-N) was directly distilled by an automatic Kjeldahl nitrogen analyzer. Determination of ether extract concentration was by the ether extraction method (<u>AOAC 2011</u>). Crude ash concentration was determined by burning at 550 °C for 3 h and water-soluble carbohydrate (WSC) concentration by the anthronesulfuric acid method (<u>Murphy 1958</u>). Buffering capacity was determined by hydrochloric acid and sodium hydroxide titration (<u>Playne and McDonald 2010</u>), while crude fiber, neutral detergent fiber (NDF) and acid detergent fiber (ADF) were determined by the filter bag method (<u>Van Soest et al. 1991</u>). Nitrogen free extract was calculated based upon the concentrations of CP, crude fiber, ether extract and crude ash. Relative feed value (RFV) was calculated based on concentrations of ADF and NDF (<u>Rohweder et al. 1978</u>).

The numbers of lactic acid bacteria (LAB), aerobic bacteria, yeasts and molds were counted by culturing on de Man Rogosa Sharpe agar, nutrient agar and potato dextrose agar, respectively. The lactic acid bacteria were cultured for 2–3 d at 37 °C under anaerobic conditions (YQX II anaerobics box, Shanghai Xinmiao Medical Device Manufacturing Co. Ltd, Shanghai, China). Aerobic bacteria, yeasts and molds were cultured under aerobic conditions at 37 °C for 3–4 days (Liu et al. 2013).

After the silage bags were opened, 20 g of the mixed silage was placed in a polyethylene plastic bag, to which 80 mL of distilled water was added before sealing. After soaking at 4 °C for 18 h, the contents were filtered and the pH of the extract was determined using a pH meter. The concentrations of lactic acid, acetic acid, propionic acid and butyric acid were determined by high performance liquid chromatography (column: Sodex RS Pak KC-811, Showa Denko KK, Kawasaki, Japan), and the operating conditions were the same as in the study by Xie et al. (2012).

Statistical analysis

Data from Experiment 1 were analyzed by 2-way analysis of variance to evaluate the effects of seeding rate, fertilizing time and their interaction on the yield, nutrient composition and silage fermentation characteristics of WCW. In Experiment 2, data were analyzed by a one-way analysis of variance. The means were compared for significance by Duncan's multiple range method (SPSS 17.0 for Windows; SPSS Inc., Chicago, IL, USA).

Results

Experiment 1

Plant height, tiller number, yield and relative feed value. Seeding rate, fertilizing time and their interaction had no significant effects on plant height or tiller number per plant (P>0.05) (Table 4). However, increasing seeding rate significantly (P<0.01) increased DM and CP yields of wheat forage but reduced relative feed value (P<0.05). Timing of the second application of fertilizer significantly (P<0.05) affected only CP yield with yields from application at jointing stage exceeding that at heading.

Chemical composition. Mean DM concentration in fresh wheat forage was 235 g/kg fresh material. There was no significant interaction between time of fertilizer application and seeding rate for concentrations of CP, crude fiber, ether extract, crude ash, nitrogen-free extract, NDF and ADF (P>0.05), but there was significant interaction for WSC concentration and buffering capacity (P<0.05) (Table 5). In general, WSC concentration and buffering capacity were higher (P<0.05) when the second fertilizer application was made at jointing rather than at heading. With the increase in seeding rate, NDF and ADF concentrations in WCW tended to increase, but CP concentration tended to decrease. Regardless of whether the second fertilizer application was made at the jointing or heading stage, seeding rate had no significant effect on CP (range 88.4-97.4 g/kg DM), ether extract and NDF (range 608-660 g/kg DM) concentrations (P>0.05). While populations of yeast, molds and aerobic bacteria were unaffected by treatment, LAB populations were consistently higher at the intermediate fertilizer level (P<0.05) (Table 5).

Silage fermentation characteristics. All silages had pH between 3.64 and 3.94 with no consistent pattern between the treatments (Table 6). Concentrations of organic acids in the silages had the following ranges: lactic acid – 14.1–21.4 g/kg DM; acetic acid – 1.08–1.56 g/kg DM; butyric acid – 0.88–2.14 g/kg DM; and propionic acid – 1.04–2.68 g/kg DM, with significant differences between treatments but no consistent pattern over the various treatments. NH₃-N concentration ranged from 141 to 172 g N/kg total N, again with differences between treatments but no consistent pattern.

Experiment 2

Plant height, tiller number, yield and relative feed value. Plant height was significantly (P<0.05) affected by fertilizer type with urea>compound fertilizer>urea + compound fertilizer (Table 7). However, tiller number/plant (mean 1.75), DM yield (mean 9.25 t/ha) and CP yield (mean 1.0 t/ha) were not affected by fertilizer type (P>0.05). Relative feed value varied with fertilizer type, being higher with compound fertilizer alone than with the other fertilizers (113 vs. 103; Table 7) (P<0.05).

Chemical and microbial composition. DM concentration of fresh forage was affected by fertilizer type, being highest for urea (391 g/kg FM) and lowest for compound fertilizer alone (338 g/kg FM) (P<0.05) (Table 8). However, fertilizer type had no effect (P>0.05) on concentrations of CP (mean 108 g/kg DM), ADF (mean 313 g/kg DM) and WSC (mean 105 g/kg DM) in forage or pH (mean 5.55). On the other hand, fertilizer type affected NDF concentration (urea and urea + compound fertilizer>compound fertilizer) and buffering capacity (compound fertilizer>urea>urea + compound fertilizer). Fertilizer type had no effect on numbers of lactic acid bacteria, aerobic bacteria, yeasts or molds in fresh forage (P>0.05).

Silage fermentation characteristics. Fertilizer type had significant effects on the pH value and concentrations of organic acids (except propionic acid) and NH₃-N in WCW silage (P<0.05) (Table 9). The pH value for silages from both urea and compound fertilizer alone exceeded that from the combined fertilizer (4.21 vs. 4.05; Table 9). While lactic acid concentration for silage from the urea + compound fertilizer treatment was greater than that from the other treatments, acetic acid concentration was higher for silage from the urea treatment than from the 2 treatments containing compound fertilizer (P<0.05). Propionic acid concentration in the various silages did not differ (P>0.05), while butyric acid concentration in silages followed the order: urea>compound fertilizer>urea + compound fertilizer (P<0.05). NH₃-N concentration was greater for silage from the urea treatment than from the other 2 treatments (P<0.05).

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| Parameter | Jointing stage | | | Heading stage | | | Significance | | |
|---------------------|--------------------|--------------------|--------------------|--------------------|-----------------|-----------------|------------------|--------------|-------------|
| | S320 | S385 | S450 | S320 | S385 | S450 | Fertilizing time | Seeding rate | Interaction |
| Plant height (cm) | 85.2 ± 2.63 | 87.8 ± 2.36 | 91.6 ± 1.72 | 87.5 ± 1.28 | 86.6 ± 1.12 | 87.3 ± 1.33 | NS | NS | NS |
| Tiller number/plant | 2.13 ± 0.20 | 1.96 ± 0.14 | 1.63 ± 0.12 | 1.88 ± 0.15 | 1.67 ± 0.13 | 1.83 ± 0.14 | NS | NS | NS |
| DM yield (t/ha) | $7.87 \pm 0.37c$ | $9.41 \pm 0.39 ab$ | $10.69 \pm 0.21a$ | $8.21 \pm 0.37 bc$ | $8.04\pm0.67bc$ | $10.30\pm0.56a$ | NS | ** | NS |
| CP yield (t/ha) | $0.77 \pm 0.04 bc$ | $0.88 \pm 0.04 ab$ | $0.98\pm0.04a$ | $0.75 \pm 0.03 bc$ | $0.71\pm0.06c$ | $0.92\pm0.04a$ | * | ** | NS |
| Relative feed value | $93.3 \pm 1.42 ab$ | $91.0\pm2.21b$ | $89.8 \pm 1.08 bc$ | $97.3\pm2.62a$ | $92.5\pm2.36ab$ | $84.7\pm0.83c$ | NS | * | NS |

Table 4. Effects of seeding rate and fertilizing time on forage and crude protein yields, plant height and tiller number (± SD) of wheat and relative feed value.

Means within a row with different letters differ at P<0.05. S₃₂₀, seeding rate of 320 kg/ha; S₃₈₅, seeding rate of 385 kg/ha; S₄₅₀, seeding rate of 450 kg/ha.

Table 5. Effects of seeding rate and fertilizing time on chemical and microbial composition (\pm SD) of wheat forage.

| Items | | Jointing stage | | | Heading stage | | S | Significance | |
|--------------------------------------|-------------------|------------------|-------------------|-------------------|------------------|--------------------|------------------|--------------|-------------|
| - | S320 | S385 | S450 | S ₃₂₀ | S385 | S450 | Fertilizing time | Seeding rate | Interaction |
| Crude protein (g/kg DM) | 97.4 ± 0.53 | 93.4 ± 0.56 | 91.5 ± 1.79 | 91.5 ± 0.28 | 88.4 ± 0.42 | 89.2 ± 1.13 | NS | NS | NS |
| Crude fiber (g/kg DM) | 330 ± 7.3 | 347 ± 3.2 | 358 ± 12.0 | 318 ± 6.1 | 329 ± 12.7 | 336 ± 11.1 | NS | NS | NS |
| Ether extract (g/kg DM) | 26.2 ± 0.43 | 25.8 ± 0.07 | 26.0 ± 0.74 | 29.6 ± 0.71 | 27.1 ± 0.33 | 29.6 ± 1.01 | NS | NS | NS |
| Crude ash (g/kg DM) | $82.9 \pm 1.71 a$ | $72.7\pm3.00b$ | $70.6 \pm 1.94 b$ | $80.2 \pm 1.23a$ | $66.9\pm0.69b$ | $73.49 \pm 2.32 b$ | NS | ** | NS |
| Nitrogen free extract (g/kg DM) | 464 ± 9.9abc | $462 \pm 8.3 bc$ | $452 \pm 17.9c$ | 481 ± 17.6 ab | 488 ± 6.1a | 476 ± 11.4 ab | ** | NS | NS |
| Neutral detergent fiber (g/kg DM) | $616\pm8.2b$ | $627\pm8.1b$ | $629\pm0.4b$ | $608\pm8.9b$ | $633 \pm 10.3 b$ | $660 \pm 2.5a$ | NS | ** | NS |
| Acid detergent fiber (g/kg DM) | $349 \pm 2.5 abc$ | $354 \pm 9.9ab$ | $362 \pm 9.9ab$ | $325 \pm 12.1c$ | $334 \pm 7.8 bc$ | $370 \pm 4.7a$ | NS | * | NS |
| Water-soluble carbohydrate (g/kg DM) | $114 \pm 0.5a$ | $110\pm0.8ab$ | 111 ± 2.0 ab | $99.6 \pm 2.51 d$ | $106 \pm 1.8 bc$ | $101 \pm 2.5 cd$ | ** | NS | ** |
| pH | $5.96\pm0.06a$ | $5.92\pm0.02a$ | $5.95\pm0.05a$ | $5.64\pm0.06b$ | $5.92\pm0.02a$ | $5.87\pm0.03a$ | NS | ** | NS |
| Buffering capacity (mE/kg DM) | $271 \pm 2.6b$ | $320 \pm 3.0a$ | $258\pm5.0b$ | $224\pm2.7d$ | $242 \pm 276c$ | $241 \pm 7.7c$ | ** | ** | ** |
| Lactic acid bacteria (lg cfu/g FM) | $5.98 \pm 0.24 b$ | $6.87\pm0.07a$ | $5.92\pm0.17b$ | $5.45\pm0.15b$ | $6.68\pm0.03a$ | $5.57\pm0.20b$ | NS | ** | NS |
| Aerobic bacteria (lg cfu/g FM) | 8.45 ± 0.03 | 8.49 ± 0.02 | 8.43 ± 0.03 | 8.37 ± 0.09 | 8.46 ± 0.07 | 8.43 ± 0.05 | NS | NS | NS |
| Yeasts (lg cfu/g FM) | 6.49 ± 0.11 | 6.51 ± 0.12 | 6.47 ± 0.18 | 6.55 ± 0.04 | 6.40 ± 0.07 | 6.42 ± 0.11 | NS | NS | NS |
| Molds (lg cfu/g FM) | 4.96 ± 0.14 | 4.90 ± 0.10 | 4.90 ± 0.10 | 4.70 ± 0.01 | 4.80 ± 0.10 | 4.86 ± 0.16 | NS | NS | NS |

Means within a row with different letters differ at P<0.05. S_{320} , seeding rate of 320 kg/ha; S_{385} , seeding rate of 385 kg/ha; S_{450} , seeding rate of 450 kg/ha. FM, fresh matter; DM, dry matter; lg, denary logarithm of the numbers; cfu, colony-forming unit.

Table 6. Effects of seeding rate and fertilizing time on the fermentation quality of wheat silage.

| 0 | | 0 | | | 0 | | | | |
|------------------------------|------------------|------------------|------------------|------------------|--------------------|--------------------|------------------|--------------|-------------|
| Items | | Jointing stage | | | Heading stage | | | Significance | |
| | S ₃₂₀ | S ₃₈₅ | S_{450} | S ₃₂₀ | S ₃₈₅ | S_{450} | Fertilizing time | Seeding rate | Interaction |
| pH | $3.75 \pm 0.00c$ | $3.85\pm0.02b$ | $3.64 \pm 0.01d$ | $3.68 \pm 0.01d$ | $3.83\pm0.01b$ | $3.94 \pm 0.03a$ | ** | ** | ** |
| Lactic acid (g/kg DM) | $19.4\pm0.14b$ | $16.6 \pm 0.30c$ | $21.4\pm0.87a$ | $18.5 \pm 0.23b$ | $15.5 \pm 0.28 cd$ | $14.1\pm0.45d$ | ** | ** | ** |
| Acetic acid (g/kg DM) | $1.18\pm0.01 bc$ | $1.56 \pm 0.04a$ | $1.33 \pm 0.12b$ | $1.09 \pm 0.01d$ | $1.24 \pm 0.02 bc$ | 1.08 ± 0.11 cd | ** | ** | NS |
| Propionic acid (g/kg DM) | $1.69\pm0.09b$ | $2.68\pm0.04a$ | $2.58\pm0.17a$ | $1.04 \pm 0.16c$ | $2.38\pm0.19a$ | $2.51 \pm 0.22a$ | * | ** | NS |
| Butyric acid (g/kg DM) | $1.28\pm0.11b$ | $1.31 \pm 0.09b$ | $1.47 \pm 0.12b$ | $0.88 \pm 0.05c$ | $1.57\pm0.07b$ | $2.14 \pm 0.03a$ | NS | ** | ** |
| NH ₃ -N (g/kg TN) | $172 \pm 1.18a$ | $163 \pm 3.03b$ | $141 \pm 1.22d$ | $133 \pm 2.24e$ | $153 \pm 2.10c$ | $157 \pm 3.18 bc$ | ** | ** | ** |

Means within a row with different letters differ at P<0.05. S₃₂₀, seeding rate of 320 kg/ha; S₃₈₅, seeding rate of 385 kg/ha; S₄₅₀, seeding rate of 450 kg/ha. TN, total nitrogen.

| Table 7. Effects of fertilizer type on yield and relative feed value of wheat for |
|---|
|---|

| Urea | Compound fertilizer | Urea + compound fertilizer |
|------------------|---|---|
| $86.8 \pm 1.04a$ | $83.8 \pm 1.14 b$ | $82.3 \pm 1.08c$ |
| 1.67 ± 0.14 | 1.79 ± 0.15 | 1.79 ± 0.17 |
| 9.34 ± 0.69 | 9.22 ± 0.74 | 9.19 ± 0.88 |
| 0.99 ± 0.07 | 1.00 ± 0.80 | 1.00 ± 0.10 |
| $103 \pm 1.45 b$ | $113 \pm 0.25a$ | $103 \pm 0.82b$ |
| | $Urea \\ 86.8 \pm 1.04a \\ 1.67 \pm 0.14 \\ 9.34 \pm 0.69 \\ 0.99 \pm 0.07 \\ 103 \pm 1.45b \\$ | UreaCompound fertilizer $86.8 \pm 1.04a$ $83.8 \pm 1.14b$ 1.67 ± 0.14 1.79 ± 0.15 9.34 ± 0.69 9.22 ± 0.74 0.99 ± 0.07 1.00 ± 0.80 $103 \pm 1.45b$ $113 \pm 0.25a$ |

Means within a row with different letters differ at P<0.05

| | Table 8. | Effects of fertilizer type | on chemical and microorganisi | m composition $(\pm SD)$ |) of wheat forage |
|--|----------|----------------------------|-------------------------------|--------------------------|-------------------|
|--|----------|----------------------------|-------------------------------|--------------------------|-------------------|

| Parameter | Urea | Compound fertilizer | Urea + compound fertilizer |
|--------------------------------------|-------------------|---------------------|----------------------------|
| Dry matter (g/kg FM) | 393 ± 0.5a | $338 \pm 3.8c$ | $351 \pm 4.3b$ |
| Crude protein (g/kg DM) | 106.5 ± 1.25 | 108.1 ± 2.70 | 109.1 ± 0.40 |
| Crude fiber (g/kg DM) | $301 \pm 6.4b$ | $340 \pm 2.8a$ | $324 \pm 6.7a$ |
| Ether extract (g/kg DM) | $25.1 \pm 1.50 b$ | $32.1 \pm 0.70a$ | $26.6\pm0.75b$ |
| Crude ash (g/kg DM) | 50.6 ± 1.39 | 54.8 ± 0.87 | 53.4 ± 1.29 |
| Nitrogen free extract (g/kg DM) | $517 \pm 6.0a$ | $465 \pm 16.9 b$ | $487\pm 6.4b$ |
| Neutral detergent fiber (g/kg DM) | 579 ± 5.3a | $537 \pm 10.7b$ | $580 \pm 1.5a$ |
| Acid detergent fiber (g/kg DM) | 322 ± 5.3 | 301 ± 8.6 | 315 ± 7.5 |
| Water-soluble carbohydrate (g/kg DM) | 103 ± 1.6 | 111 ± 6.0 | 102 ± 1.1 |
| рН | 5.51 ± 0.11 | 5.45 ± 0.03 | 5.70 ± 0.21 |
| Buffering capacity (mE/kg DM) | $183 \pm 2.9b$ | $219 \pm 3.4a$ | $161 \pm 9.5c$ |
| Lactic acid bacteria (lg cfu/g FM) | 5.94 ± 0.13 | 6.05 ± 0.03 | 6.23 ± 0.05 |
| Aerobic bacteria (lg cfu/g FM) | 8.23 ± 0.02 | 8.35 ± 0.01 | 8.37 ± 0.05 |
| Yeasts (lg cfu/g FM) | 6.54 ± 0.06 | 6.79 ± 0.08 | 6.74 ± 0.09 |
| Molds (lg cfu/g FM) | 5.00 ± 0.17 | 4.80 ± 0.10 | 4.90 ± 0.10 |

Means within a row with different letters differ at P<0.05. FM, fresh matter; lg, denary logarithm of the numbers; cfu, colony-forming units.

| Table 9. Effects of fertilizer type on the fermentation quality $(\pm SD)$ of wheat sil |
|--|
|--|

| 21 | 1 2 1 | 6 | |
|------------------------------|------------------|---------------------|----------------------------|
| Parameter | Urea | Compound fertilizer | Urea + compound fertilizer |
| pH | $4.24 \pm 0.03a$ | $4.18 \pm 0.05a$ | $4.05\pm0.01b$ |
| Lactic acid (g/kg DM) | $12.03\pm0.41b$ | $11.88\pm0.27b$ | $14.09 \pm 0.70a$ |
| Acetic acid (g/kg DM) | $1.62 \pm 0.17a$ | $1.17 \pm 0.14b$ | $1.15 \pm 0.02b$ |
| Propionic acid (g/kg DM) | 2.61 ± 0.33 | 2.36 ± 0.22 | 2.26 ± 0.03 |
| Butyric acid (g/kg DM) | $2.87 \pm 0.11a$ | $2.48 \pm 0.12 b$ | $1.01 \pm 0.02c$ |
| NH ₃ -N (g/kg TN) | $150 \pm 14.2a$ | $124 \pm 3.4b$ | $132 \pm 11.9b$ |
| | | | |

Means within a row with different letters differ at P<0.05. TN, total nitrogen.

Discussion

Both seeding rate and timing of fertilizer application are considered important management strategies affecting crop production, and an optimal seeding rate can achieve a balance between yield of wheat forage and cost of seed. In general, increasing seeding rates results in higher yield (Counce et al. 1992; Jia et al. 2018).

In this study, both DM and CP yields of forage planted at 450 kg seed/ha (S₄₅₀) were significantly higher than that of S₃₂₀ (P<0.05), which supports the statement above. In general, high seeding rate of crops exacerbates the competition among plants for critical resources such as water, nutrients and light (<u>Xue et al. 2016</u>), so accumulation of DM per plant can be reduced, but the higher plant population more than makes up for the reduction in DM yield per plant, thus increasing yield (<u>Liu</u> et al. 2011). Plants at the higher seeding rate in our study possibly intercepted more incident light, thus resulting in greater DM accumulation, which is consistent with the results of Arduini et al. (2006).

Tran and Tremblay (2000) found that applying fertilizer at the heading stage promoted the growth of wheat during the reproductive period, reduced the effects of inefficient tillering and increased the nitrogen concentration in grain. In our study, time of applying the second application of fertilizer had no significant effect on most of the parameters measured, suggesting that timing of fertilizer application in this case was not critical.

With the increase in seeding rate, concentrations of crude fiber, NDF and ADF in wheat forage tended to increase in this study but differences failed to reach significance. This suggests that the nutritive value of WCW would tend to decrease at higher seeding rates as was shown by a trend of lowering relative feed value as seeding rate increased.

When wheat was fertilized at the heading stage, the WSC concentration in the forage tended to decrease, compared with wheat fertilized at the jointing stage. From the perspective of silage production, higher WSC concentration can promote lactic acid fermentation and improve silage fermentation quality. Lactic acid and acetic acid production in silages in Experiment 1 showed no consistent pattern across treatments but pH of all silages was in the range 3.64-3.94, indicating good quality silage, which was reinforced by the low concentrations of butyric acid (0.88-2.14 g/kg DM). When fertilizer application occurred at the jointing stage, the NH₃-N concentration in the silage decreased significantly with increase of seeding rate, indicating that protein decomposition of silage was low under high seeding rate. However, when fertilizer was applied at the heading stage, the silage fermentation quality of wheat forage decreased with increasing seeding rate. Generally, the production of acetic acid is dominated by Enterobacter, Enterococcus and Clostridium, which are also the bacteria that decompose amino acids to produce NH₃-N. Enterobacter also dominates the production of NPN, degrading protein by secreting carboxypeptidase (Li 2018).

Nitrogen from urea is released rapidly in the early stages after application to the soil when the release rate can exceed the crop demand, which can result in insufficient N supply in the later stages of crop growth. In this study, substituting compound fertilizer for urea or combining urea and compound fertilizer, resulted in no significant change in DM yield of WCW, indicating that the N component was the over-riding factor determining growth of the wheat and losses of N from volatilization of urea were not a significant issue. Increased quantities of phosphate (P) and potassium (K) obviously had no effect on growth of the wheat. Beauregard et al. (2010) suggested that applying P₂O₅ could directly or indirectly change chemical, physical and biological the characteristics of soil, increase soil P availability and increase the CP concentration of forage without having any significant effect on forage yield. Given the available P and K levels in the soil where the study was conducted, it is not surprising that there were no DM yield responses to compound fertilizer over that with urea application. The application of compound fertilizer improved the relative feed value of wheat, which was a function of a significant increase in ether extract and a significant reduction in NDF concentration in forage from this treatment. Berg et al. (2007) found that application of phosphate fertilizer reduced NDF and ADF concentrations in forage.

NH₃-N, acetic acid and butyric acid concentrations in silage from the urea treatment were higher than those in silages from compound fertilizer and urea + compound fertilizer treatments, which supported the results reported by Namihira et al. (2011). The wheat silage from the urea + compound fertilizer treatment had the highest lactic acid concentration and the lowest butyric acid concentration in the 3 fertilizer treatments, possibly because the lower buffering capacity accelerated the decrease in pH and promoted the fermentation of lactic acid.

Conclusions

This study has shown that WCW has the propensity for high yields of forage of high feeding value. Under the conditions of this study, considering DM yield, nutrient composition and silage fermentation quality, a seeding rate of wheat for forage of 385 kg/ha would seem appropriate. If fertilizer application to wheat is to be split, applying a part at jointing stage would be more beneficial than that at heading stage. Compared with urea and compound fertilizer alone, applying urea with compound fertilizer did not affect the yield and nutritive value of WCW, but did improve the silage fermentation quality. These results need verification on different soil types.

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