Chemical composition, in vitro ruminal dry matter degradability and dry matter intake of some selected browse plants

Hilda Kwena Mokoboki, Amenda Nthabiseng Sebola, Khuliso Emmanuel Ravhuhali* and Lindani Nhlane

Abstract: Many browse plants are undervalued due to insufficient knowledge about their potential feeding value. The study was conducted to evaluate the chemical composition, in vitro ruminal dry matter degradability (DRD) and dry matter intake of Melia azedarach, Leucaena leucocephala, Searsia lancea, Moringa oleifera and Acacia hebeclada. Matured leaves were collected and dried at room temperature and then analyzed in a completely randomized design. Chemical composition, ruminal dry matter degradability and dry matter intake were determined and estimated, respectively. The crude protein (CP) content ranged from 108.0 to 258.3 g/kg DM and neutral detergent fiber from 306.1 to 492.7 g/kg DM. In vitro dry matter degradability at all incubation periods - for M. oleifera was significantly higher than those of other browse plants. There was a negative correlation between metabolizable energy and fiber content and a positive correlation to crude protein content. All browse species had CP values of above 10% which is above the minimum required in diet for adequate digestive activities. M. oleifera, L. leucocephala and M. azedarach leaves had the...
highest metabolizable energy, dry matter intake, total digestible nutrients, relative feed value and dry matter degradability compared to other browse plants. The results from the study suggest that *M. oleifera, M. azedarach* and *L. leucocephala* can be recommended to be used as a supplement during dry season and in case of low-quality forage feeding.

**Subjects:** Agriculture; Agriculture and Food; Nutrition

**Keywords:** Chemical composition; browse; DM intake; in vitro degradability

### 1. Introduction

A lot of research has been done on the importance of forage browse trees (Ogunbosoye, Tona, & Otukoya, 2015) and legumes (Le Houerou, 1980) as sources of valuable protein for ruminants (Norton, 1982). Browse species play a significant role in ruminant animals through their contribution to enhanced nutritive value of the animal diet, biological nitrogen (N) fixation (Humphreys, 1995) provide vitamins, minerals and protein which are lacking in mature natural pasture especially during the dry season (Skerman, Cameron, & Riveros, 1988). It has been suggested that they can be used as protein supplements in ruminant feeding because of their high protein content (Skerman et al., 1988).

Protein supplementation from these sources could enhance carbohydrate fermentation in animals on low-quality roughages (Molina-Alcaide, Weisbjerg, & Hvelplund, 1996). There is an increasing interest in the rational utilization of potential livestock feed resources such as browse species. Communal farmers cannot afford to procure protein supplementary feeds due to their high cost (Evitayani, Wary, Fariani, Ichinohe, Abdulrazak, and Fujihara, 2004). Browse plants such as *Melia azedarach*, *Acacia hebeclada*, *Moringa oleifera*, *Leucaena leucocephala* and *Searsia lancea*, which are available in the country, can be used as a cheap protein supplement. Browse plants are also useful sources of animal feeds, as these plants remain green during the dry season and also provide vegetation with better nutritive value than other annual grass and herbaceous species that become withered (Aregawi, Melaku, and Nigatu, 2008). These browse plants can prevent desertification by mitigating the effects of droughts, allowing soil fixation and enhancing the restoration of the vegetation (Robles, Ruiz-Mirazo, Ramos, & Gonzalez-Regollar, 2008). There is a need for an adequate understanding of the nutrient availability, both in the rumen and postruminally, from these plant species in order to properly balance their use in ruminant nutrition. Many browse plants have been undervalued mainly because of insufficient knowledge about their potential feeding value. Some of the browse plants studied herein are consumed to a certain degree by small ruminants grazing and are indispensable as feeds for herbivores when production systems are based on grazing rangelands (Papanastasis, Yialkoulaki, Decandia, & Dini-Papanastasi, 2008). The objective of the present study was to assess the chemical composition, in vitro ruminal dry matter degradability and dry matter intake prediction of the selected browse plants.

### 2. Materials and methods

#### 2.1. Study site

The study was conducted at the Molelwane Farm situated 6 km west of the North West University, South Africa. The geographical coordinates of Molelwane are 25°48′00″S and 25°38′21″E. The area has summer months from August to March with temperatures ranging from 22°C to 35°C and a long-term average annual rainfall of 450 mm. The province has winter months from May to July, with sunny dry days and chilly nights with average minimum and maximum temperatures of 2°C and 20°C respectively.

#### 2.2. Forages

Browse trees replicated 10 times from each of the five (*M. azedarach, A. hebeclada, M. oleifera, L. leucocephala* and *S. lancea*) browse species were randomly selected and marked for harvesting.
during the summer. The leaves were collected by handpicking and air-dried at room temperature in a well-ventilated laboratory. Samples were ground in a hammer mill to pass a 2-mm screen for both chemical and rumen degradability analyses.

2.3. Chemical analysis
Moisture and dry matter contents were determined after drying samples in an oven at 105°C to constant weight. A muffle furnace set at 600°C was used to determine organic matter (OM) content by ashing the dried samples for 6 h. The loss in weight was measured as organic matter and the residue as ash. The standard macro-Kjeldahl method (Association Of Analytical Chemists, 2005, method no. 978.04) was used to determine the total nitrogen content and was converted into CP by multiplying percentage of N content by a factor 6.25. Neutral detergent fiber (NDF) and ADF were determined according to Van Soest, Robertson, and Lewis (1991). Acid detergent lignin (ADL) was determined by placing dried ADF bags/samples in sufficient quantity of 72% H$_2$SO$_4$. The formula used to predict total digestible nutrients (TDN) was $82.38 - (0.7515 \times ADF)$ as described by Bath and Marble (1989). Relative feed value (RFV) was calculated from the estimates of dry matter digestibility (DMD) (Jerenyama & Garcia, 2004) and dry matter intake (DMI) (Mertens (2002)): DMD % = 88.9 - (0.779 × %ADF), DMI = 1.2 × body weight/NDF%, RFV = (%DDM x %DMI)/1.29. Dry matter digestibility values were then used to estimate digestible energy (DE, kcal/kg) using the regression equation reported by Fonnesbeck, Clark, Garret, and Speth (1984): $DE (Mcal/kg) = 0.27 + 0.0428 \times DMD%$. Then, DE values were converted to ME using the formula reported by Khalil, Sawaya, and Hyder (1986): ME (Mcal/kg) = 0.821 × DE (Mcal/kg). Estimated predicted potential daily DMI was calculated according to Mertens (2002) using the formula

$$DMI = \frac{1.2 \times \text{body weight}}{NDF\%}$$

where body weight is equivalent to an estimated livestock unit metabolic weight of 450 kg.

2.4. Rumen degradability
The in vitro ruminal DM degradability of leaf samples was determined using the DaisyII incubator consisting of a thermostatic chamber (39°C) with four rotating jars according to ANKOM Technology Method 3 for in vitro true digestibility (ANKOM Technology Corp., Fairport, NY). In brief, ground samples (2 mm) were weighed in ANKOM F57 bags (0.45–0.5 g), heat sealed and placed in the digestion jars filled with pre-warmed buffer solutions and rumen inoculum collected from rumen—fisculated bonsmara cow. Filter bags were withdrawn at 6, 12, 24, 36, 48 and 72 h after inoculation. In vitro ruminal DM degradability was determined using the following formula:

$$\% \text{IVTD} (\text{DM basis}) = \frac{100 - (W3 - (W1 \times C1))}{W2 \times DM} \times 100$$

where $W1 =$ bag tare weight, $W2 =$ sample weight, $W3 =$ final bag weight after in vitro treatment, and $C1 =$ blank bag correction factor (final oven-dried weight ÷ original blank bag weight).

2.5. Ethical consideration
The fistulated animal, which was cared for according to institutional as well as Federation of Animal Science Societies guidelines (FASS, 2010) for the care and use of agricultural animals in research and teaching, was used for the in vitro degradability trial experiment. Ethical approval was obtained from the North West University Animal Research Ethics Committee; approval number NWU-00126-13-A9.

2.6. Statistical analysis
All data were analyzed by ANOVA using the one-way analysis of variance of GLM procedure of SAS (2010) for completely randomized design according to the following statistical model:

$$Y_{ij} = \mu + AS + E_{ij}$$
where $Y_{ij}$ = measured variable, $\mu$ = mean, $AS$ = effect of treatment and $E_{ij}$ = random error, where significant variation was detected and comparison of treatment means was carried out using least square means.

Regression analyses were used to determine the relationship between chemical composition parameters and in vitro ruminal degradability at 36 and 72 h. The correlation coefficient ($r$) between dry matter degradability, chemical composition and estimated parameters was also used.

3. Results
Chemical composition variables except hemicellulose were significantly influenced ($P < 0.05$) by browse plant species (Table 1). Melia azedarach, L. leucocephala and M. oleifera leaves had the highest ($P < 0.05$) crude protein content and low fiber content compared to S. lancea and A. hebeclada.

Predicted dry matter intake and relative feed value were significantly higher ($P < 0.05$) for L. leucocephala, M. azedarach and M. oleifera compared to A. hebeclada and S. lancea (Table 2).

In vitro dry matter degradability at all incubation periods for M oleifera was significantly higher ($P < 0.05$) than those of other browse plants.

There was a positive correlation ($P < 0.05$) between dry matter degradability from 6 to 36 h incubation period (Table 3) and hemicellulose, dry matter intake, relative feed value and crude protein content.

### Table 1. Chemical composition (g/kg DM) of selected browse plants

| Browse species | DM g/kg | OM g/kg | CP g/kg | NDF g/kg | ADF g/kg | ADL g/kg | Cellulose g/kg | Hemicellulose g/kg |
|---------------|---------|---------|---------|----------|--------|---------|---------------|------------------|
| M. azedarach  | 938.0   | 859.1   | 206.7   | 320.8    | 266.9  | 118.0   | 148.9         | 53.9             |
| S. lancea     | 937.7   | 907.9   | 108.0   | 432.0    | 313.1  | 99.7    | 213.4         | 118.9            |
| A. hebeclada  | 949.1   | 907.8   | 127.6   | 492.7    | 391.2  | 226.7   | 164.6         | 101.5            |
| L. leucocephala| 928.2  | 863.0   | 186.6   | 306.1    | 212.3  | 112.6   | 99.7          | 93.8             |
| M. oleifera   | 957.4   | 891.8   | 258.3   | 309.3    | 221.9  | 126.0   | 95.9          | 87.4             |
| SE            | 2.9     | 3.0     | 0.0     | 15.0     | 15.3   | 10.0    | 7.8           | 21.5             |

$^{abcd}$ Means within column with no common superscripts differ significantly ($P < 0.05$).
SE = standard error.
DM = dry matter, OM = organic matter, CP = crude protein, NDF = neutral detergent fiber, ADF = acid detergent fiber, ADL = acid detergent lignin.

### Table 2. Metabolizable energy, dry matter digestibility, dry matter intake, total digestible nutrients and relative feed value of selected browse plants

| Browse species | Metabolizable energy, MJ/kg | DMD g/kg DM | DMI g/kg DM | TDN g/kg DM | RFV g/kg DM |
|---------------|---------------------------|-------------|-------------|-------------|-------------|
| M. azedarach  | 0.54$^a$                  | 207.9$^a$   | 16.8$^a$    | 623.2$^a$   | 888.7$^a$   |
| S. lancea     | 0.51$^b$                  | 243.9$^b$   | 12.5$^b$    | 588.5$^b$   | 625.4$^b$   |
| A. hebeclada  | 0.46$^c$                  | 304.8$^c$   | 10.9$^c$    | 529.7$^c$   | 496.5$^c$   |
| L. leucocephala| 0.55$^a$                | 193.9$^a$   | 17.7$^a$    | 636.7$^a$   | 951.6$^a$   |
| M. oleifera   | 0.53$^a$                  | 215.4$^a$   | 17.7$^a$    | 615.9$^a$   | 921.0$^a$   |
| SE            | 0.14                      | 16.34       | 0.75        | 15.76       | 26.47       |

$^{abc}$ Means within column with no common superscripts differ significantly ($P < 0.05$).
SE = standard error.
DMD = dry matter digestibility, DMI = dry matter intake, TDN = total digestible nutrients, RFV = relative feed value.
The incubation period of dry matter degradability was not correlated (P > 0.05) to organic matter, cellulose, acid detergent lignin and neutral detergent fiber (except at 6 h incubation period) (Table 4).

Metabolizable energy, dry matter digestibility, total digestible nutrients and relative feed value were negatively correlated (P > 0.05) with organic matter, neutral detergent fiber, acid detergent fiber and acid detergent lignin whereas the same parameters were positively correlated (P < 0.05) with crude protein content (Table 5).

Dry matter degradability at 36 and 72 h incubation periods was best predicted by the equation that included crude protein content (Table 6). The regression coefficient values ranged from 0.005 to 0.868 for 36 h incubation period and 0.021 to 0.803 for 72 h incubation period.

4. Discussion

Wide variations were observed in chemical composition, dry matter degradability and dry matter intake of different browse plants. This is in agreement with the report by Duguna, Tonye, Kannmegne, and Enoch (1994) that multipurpose tree leaves vary widely in their chemical composition. The crude protein content in the leaves of four browse plants except S. lancea was in excess of recommended minimum requirements for growth (113 g/kg DM) and lactation (120 g/kg DM) in ruminant animals (ARC, 1984).

According to Paterson, Cohran, and Klopfenstein (1996), feedstuffs with CP lower than 70 mg/g DM require a supplementation of nitrogen to improve their digestion and ingestion by the ruminants. The crude protein content of all selected browse plants was above 80 g/kg DM, the reported minimum required in diet for adequate digestive activities (Orskov, 1982). The high crude protein content in the leaves of four browse plants except S. lancea was in excess of recommended minimum requirements for growth (113 g/kg DM) and lactation (120 g/kg DM) in ruminant animals (ARC, 1984).

Table 3. In vitro dry matter degradability of some selected browse plants incubated with rumen fluid

| Species          | DMD0 | DMD6 | DMD12 | DMD24 | DMD36 | DMD48 | DMD72 |
|------------------|------|------|-------|-------|-------|-------|-------|
| M. azedarach     | 139.6 | 251.3 | 385.7  | 430.2  | 646.6  | 593.8 | 591.6  |
| S. lancea        | 200.8 | 248.9 | 238.4  | 260.6  | 351.0  | 325.7 | 359.8  |
| A. hebeclada     | 115.3 | 227.0 | 300.1  | 392.7  | 556.8  | 551.2 | 560.1  |
| L. leucocephala  | 92.4  | 263.7 | 332.9  | 405.7  | 568.6  | 499.2 | 548.5  |
| M. oleifera      | 236.9 | 397.9 | 495.3  | 595.1  | 845.4  | 746.6 | 791.2  |
| SE               | 26.3  | 16.0  | 15.4   | 10.5   | 19.7   | 13.9  | 13.0   |

abcd Means within column with no common superscripts differ significantly (P < 0.05).
SE. Standard error.

Table 4. Correlation coefficient (r) between dry matter degradability, chemical composition and estimated parameters

| Incubation period (h) | Chemical constituents |
|-----------------------|-----------------------|
|                       | OM | CP | NDF | ADF | CELLU | ADL | HEMI | DMI | RFV |
| 0                     | 0.40NS | -0.09NS | -0.11NS | 0.92NS | 0.08NS | -0.24NS | 0.18NS | 0.07NS | 0.08NS |
| 6                     | -0.16NS | 0.54 | -0.54 | -0.65 | 0.08NS | -0.34NS | 0.64 | 0.53 | 0.58 |
| 12                    | -0.31NS | 0.67 | -0.34NS | -0.73 | -0.42NS | -0.09NS | 0.86 | 0.70 | 0.67 |
| 24                    | -0.27NS | 0.54 | -0.27NS | -0.81 | -0.26NS | 0.07NS | 0.77 | 0.58 | 0.55 |
| 36                    | -0.34NS | 0.54 | -0.26NS | -0.77 | -0.34NS | 0.08NS | 0.80 | 0.58 | 0.55 |
| 48                    | -0.26NS | -0.44NS | -0.15NS | -0.72 | -0.32NS | 0.19NS | 0.76 | 0.48NS | 0.44NS |
| 72                    | -0.25NS | -0.51NS | -0.19NS | -0.77 | -0.31NS | 0.13NS | 0.81 | 0.55 | 0.51NS |

OM = organic matter, CP = crude protein, NDF = neutral detergent fiber, ADF = acid detergent fiber, CELLU = cellulose, ADL = acid detergent lignin, HEMI = hemicellulose, DMI = dry matter intake, RFV = relative feed value.
protein content in these plants may be an advantage to rumen microbes (especially when these browse plants have average tannins concentration) that depend on the dietary source of nitrogen to build up their body proteins (Aderinboye et al., 2016). The lower crude protein content in *S. lancea* may be due to high proportions on mature leaves in the sample. The CP level in *S. lancea* species is within the range reported by Ravhuhali (2018), and lower than those reported by Naumann, Cooper, and Muir (2016) and higher than Monegi, Mbatha, Tjelele, and Mkhize’s (2016) findings on dry leaves. The nutrient content of *L. leucocephala* leaves is lower than that of *M. oleifera* leaves which is in agreement with Kakengi et al. (2007), Foidl, Makkar, and Becker (2001) and Makkar and Becker (1997). The high crude protein content of *M. oleifera* in the current study falls within the one reported by Gadzirayi, Masamha, Mupangwa, and Washaya (2012) (25–27% DM), and this high level has been reported to be comparable to that of milk and eggs.

### Table 5. Correlation coefficient (r) among some estimated parameters and chemical composition of browse plants

| Chemical constituents | OM   | CP   | NDF  | ADF  | ADL  |
|-----------------------|------|------|------|------|------|
| Metabolizable energy  | -0.65| 0.89 | -0.69| -1.00| -0.73|
| Dry matter digestibility | -0.65| 0.89 | -0.69| -1.00| -0.73|
| Dry matter intake     | -0.72| 0.83 | -0.99| -0.61| -0.57|
| Total digestible nutrients | -0.64| 0.79 | -0.67| -1.00| -0.73|
| Relative feed value   | -0.77| 0.78 | -0.99| -0.76| -0.64|

OM = organic matter, CP = crude protein, NDF = neutral detergent fiber, ADF = acid detergent fibre, ADL = acid detergent lignin.

### Table 6. Linear regression on the prediction of in vitro dry matter degradability (g/kg DM) (DMD36 and DMD72) from chemical composition (g/kg DM for OM, CP, NDF, ADF, ADL, CEL, HEM) of different browse plants

| Factor | Y-variable | Formulae | r² | P value |
|--------|------------|----------|----|---------|
| OM     | DMD36      | Y = -2.436OM + 2752.01 | 0.106 | 0.593 |
| CP     | DMD36      | Y = 2.734CP + 108.19  | 0.868 | 0.021 |
| NDF    | DMD36      | Y = -1.232NDF + 1052.21 | 0.349 | 0.294 |
| ADF    | DMD36      | Y = -1.191ADF + 928.36 | 0.241 | 0.401 |
| ADL    | DMD36      | Y = 0.237ADL + 561.41  | 0.005 | 0.913 |
| CEL    | DMD36      | Y = -2.982CEL + 1025.02| 0.664 | 0.093 |
| HEM    | DMD36      | Y = -4.459HEM + 1000.48| 0.360 | 0.284 |
| OM     | DMD72      | Y = 1.603OM + 1990.27  | 0.062 | 0.687 |
| CP     | DMD72      | Y = 2.264CP +168.30   | 0.803 | 0.040 |
| NDF    | DMD72      | Y = -0.942NDF + 920.94| 0.275 | 0.364 |
| ADF    | DMD72      | Y = -0.914ADF + 827.37 | 0.192 | 0.461 |
| ADL    | DMD72      | Y = 0.430ADL + 511.59  | 0.021 | 0.817 |
| CEL    | DMD72      | Y = -2.565CEL + 941.25 | 0.663 | 0.094 |
| HEM    | DMD72      | Y = -3.372HEM + 877.92 | 0.278 | 0.361 |

r² = regression coefficient, P value = probability value.
OM = organic matter; CP = crude protein; NDF = neutral detergent fiber; ADF = acid detergent fiber; ADL = acid detergent lignin; CEL = cellulose; HEM = hemicellulose; DMD36 = dry matter degradability at 36 h after incubation; DMD72 = dry matter degradability at 72 h after incubation.
The crude protein content recorded in this study for *M. oleifera* of 25.8% was lower than the one reported by Oduro, Ellis, and Owusu (2008) and Sanchez-Machado, Nunez-Gastelum, Reyes-Moreno, Ramirez-Wong, and Lapenç-Cervantes (2009). The crude protein content of *M. azedarach*, *L. leucocephala* and *M. oleifera* could meet both animal’s protein and energy requirements and may be helpful in preventing diseases occurrence in animals (Brisibe, Umoren, Brisibe, Magalhaes, Ferreira, Luthria, Wu X, and Prior, 2009). The crude protein content again for these three browse plants is above 16% which is the one required for growing goats making them ideal for use as a protein supplement (Moyo, Masika, Hugo, & Muchenje, 2011). The crude protein content of *L. leucocephala* in this study was lower than 21.4% reported by Evitayani et al. (2004). However, the presence of fiber and anti-nutritional factors such as condensed tannins may reduce the bioavailability of this protein in these three browse plants.

*A. hebeclada* had the highest neutral detergent fiber value (492.7 g/kg DM) compared to other browse plants. However, it was lower than the one reported by Mokoboki, Ndlovu, Ng’Ambi, Malatje, and Nikolova (2005) (570.1 g/kg DM). *M. oleifera* neutral detergent fiber content from this study is higher than the one reported by Melesse (2011). The NDF content of *L. leucocephala* leaves from this study is lower than that reported by Gemeda and Hassen (2015) and Njidda and Nasiru (2010). The values in the current study for acid detergent fiber content of *M. oleifera* and *L. leucocephala* are lower than those reported by Njidda and Nasiru (2010).

For the five browse plants, the level of ADF ranged from 212.3 to 391.2 g/kg DM, which falls within the range where browse substrates are expected to be digestible (Belyea & Rickets, 1993) and would therefore not negatively impact on the bioavailability of CP. Research indicates that a cow will eat a daily quantity of NDF up to approximately 1.2% of its body weight (Mertens, 2002). Daily dry matter intake for the three browse species (*M. oleifera*, *L. leucocephala* and *M. azedarach*) was high because of the low neutral and acid detergent fiber content. Thus, there will be a fast rate of passage through the rumen, and the resulting gut fill will cause animals to increase their intake, though *L. leucocephala* also has issues with toxicity related to its mimosine content, which would also limit its use in ruminants not adapted to this plant species.

There was a negative correlation between acid detergent fiber content and dry matter degradability, which is in agreement with Kamalak et al. (2005) and Parissi, Papachristou, and Nastis (2005). Minson (1990) also observed the negative correlation between ADF and degradability. In contrast to this, the results reported by Repetto et al. (2003) showed that ADF has the closest relationship with DMD. Variation within browse species might have contributed to that negative correlation coefficient. Indeed, species with different chemical compositions may respond differently during degradation (Jung & Allen, 1995; Ravhuholi, Ng’ambi, & Norris, 2010). Again, negative correlation coefficient values in these browse species might be due to indigestible material such as cellulose and lignin that might have restricted DM degradation. Substrate degradability can be reduced due to more lignifications of the plant cell wall (Du, Xu, and Yao, 2016). Buxton and Redfearn (1997) highlighted that because lignin can interfere with microbial degradation of fiber polysaccharides by acting as a barrier, forage degradability can be improved by reducing the amount of lignified cells in browse species. There was a negative correlation between metabolizable energy and fiber content and a positive correlation to crude protein content which is in agreement with the findings of Tolera, Khazaal, and Ørskov (1997). Parissi et al. (2005) also reported a positive correlation between crude protein and metabolizable energy. The results of the study are also consistent with the findings of Kamalak et al. (2005). Fiber is known in affecting ME content of the substrates due to the fact that digestibility of protein will decrease when fiber intake increases and that will decrease the ME content of the diet (Baer, Rumpler, Miles, & Fahey, 1997).

5. Conclusions
The significant differences in chemical composition resulted in significant differences of dry matter degradability and dry matter intake. All browse species had CP values of above 10% which is above
the minimum required in the diet for adequate digestive activities. There was also a positive relationship between ME and CP and a negative correlation between ME and fiber content. The moderate to high crude protein content and low fiber content, along with high dry matter intake and total digestible nutrients, found in *M. oleifera*, *M. azedarach* and *L. leucocephala* suggest that these browse plants may be considered feed supplements to be used by the ruminants during feed shortage. However, feeding trial is needed to be more informative about these browse plants especially on secondary metabolites.

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The authors declare no competing interests.

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