

FARMERS' GUIDANCE FOR RECYCLING AGRICULTURAL WASTES TO PRODUCE ANIMAL FEED

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ABSTRACT

The increasing expansion of water hyacinth plant activity over year has led to the accumulation of large quantity their in the Nile River due to prohibit a lot of Nile water and light from organisms living in the Nile, and consuming Nile water. Recently, the world used water hyacinth leaves to absorb the metal ions from waste water. In this study we used this to upgrade nutritional value of some lignocellulosic materials such as rice straw and banana leaves. The lignocellulosic residues can be used without any chemical or biotechnology pretreatment (control) or can be hydrolyzed before any fermentation, then the subsequent can result in a product with high protein content as well as high digestibility compared to non added residues. In this study, we made mixture combination water hyacinth leaves to rice straw or banana leaves with different ratios (5, 10, 15, 20, 25 and 30 g water hyacinth leaves/100g rice straw or banana leaves). All treatments gave a good results, but treatment added 30 g water hyacinth leaves / 100g rice straw or banana leaves is the better than all. The increasing in protein content from 2.30%, 12.50 % (control) to 10.00%, 17.10 % and decreasing in fiber fraction especially, cellulose and lignin. Cellulose decreased from 33.30 %, 26.30% to 22.00%, 19.35% and lignin decreased from 11.60%, 8.50% and 5.95%, 5.35% in rice straw and banana leaves, respectively. *In vitro* digestibility studies indicated significantly ($P < 0.05$) higher values for treatment added 30% water hyacinth leaves in the rice straw and banana leaves, respectively, to produce feed non traditional for animals.

Pleurotus ostreatus was used to upgrade nutritional value of rice straw, banana leaves and water hyacinth leaves by using different amounts of *Pleurotus ostreatus* inoculum, in solid state fermentation technique. The chemical composition and *in vitro* digestibility of the resulted protein enriched product were determined. Cellulose, hemicellulose and lignin showed gradual decrease with increasing inoculums of *Pleurotus ostreatus*, however, *in vitro* disappearance increased.

Keywords: Rice straw, Banana leaves, Water hyacinth leaves, *Pleurotus ostreatus*, *in vitro* disappearance.

المخلص

زيادة اتساع نبات ورد النيل كل عام يؤدي إلى تراكم كميات كبيرة منه مما يعمل على فقد الكثير من مياه النيل، وكذلك يؤدي إلى نقص الأكسجين الذائب في المياه مما يهدد حياة الأسماك والكائنات البحرية. حديثاً، في العالم يستخدم نبات ورد النيل كمادة ممتصة للعناصر المعدنية الثقيلة مثل الكاديوم والنحاس والزنك والرصاص في المخلفات المائية. وفي هذه الدراسة تم استخدام ورد النيل لرفع القيمة الغذائية لبعض المخلفات اللجنوسليلوزية مثل قش الأرز وأوراق الموز.

المخلفات اللجنوسيليلوزية المستخدمة بدون اى معاملات كيميائيه او بيولوجيه (الكنترول) مقارنة بالاضافات من اوراق ورد النيل بنسب مختلفه (٥ - ١٠ - ١٥ - ٢٠ - ٢٥ و ٣٠) لكل ١٠٠ جرام من قش الارز او اوراق الموز . حيث زاد البروتين من ٢,٣٠% و ١٢,٥٠% (كنترول) الى ١٠,٠% و ١٧,١٠% ونقص فى نسيه السيليلوز والهيموسيليلوز واللجنين . حيث نقص السيليلوز من ٣٣,٣٠% و ٢٦,٣٠% الى ٢٢,٠% و ١٩,٣٥% وكذلك اللجنين من ١١,٦٠% و ٨,٥٠% الى ٥,٩٥% و ٥,٣٥% فى كل من قش الارز واوراق الموز بعد اضافته ٣٠ جرام من اوراق ورد النيل على التوالى . كذلك النسبه الهضميه كانت اعلى معدلاتها فى معاملة اضافته ٣٠ جرام من ورد النيل الى كل قش الارز واوراق الموز على التوالى لانتاج علف حيوانى غير تقليدى . كذلك استخدام عمل على رفع القيمه الغذائيه للمخلفات المستخدمه لكل من التركيب *Pleurotus ostreatus* الكيماوى لها والنسبه الهضميه .

INTRODUCTION

In Egypt, there is a limitation in the cultivated area and a high increasing in the population rate. Thus, there is a serious shortage in animal feeds, causing high negative effects on live stock production. Therefore, it is very important to use unconventional sources as animal feeds. Agriculture wastes and by products seem to overcome partially this problem.

In Egypt, there is a lack in protein feed ingredient for animal feeding. So, many studies were carried out to improve the quality of protein feed ingredient (**EL-Sayed et al., 2002**), or to use new sources of protein (**Mohi El-Din et al., 2008; Leupp et al., 2009; May et al., 2009; Shwerab et al., 2010 and Etman et al., 2011**) in ruminant rations. Agricultural crop residues are still grown dramatically causing severe pollution problems , with a production of approximate 35 million tons on DM basis per year. Traditionally, they are removed to be used as fodder, cooking fuel, fencing to be burned. Recently, return of residues into the field, such as leaving crop residues on the soil surface, incorporating them into the soil, has been popular in enhancing soil quality.

Rice straw represents an important summer crop by-product in Egypt. About 3.5 million tons of rice straw and 0.5 million tons of rice hulls are produced every year from the rice fields and rice milling process respectively. There is no practical use for these by-products, up till now, except for fuel. This paper reviews some research results relevant to the use of rice straw and rice hulls as feeds for ruminants in Egypt.

Chemical composition of banana wastes is very close to that of berseem. Banana wastes can play an important role in covering some nutrients requirements of animals (**Abd-EL-Gawad et al., 1994**) especially they are available in Egypt all over the year (**Khatab et al., 2000**). The native agricultural crop residues are slowly digested by reumines microorganisms and considered a poor fermentation substrates. Thus, physical (milling) and addition water hyacinth leaves, treatments of these substrates are necessary to disrupt the physical fine structure of cellulose as well as to remove lignin .

Water hyacinth plant was used to adsorption to several cations from waste water. The dried leaves of water hyacinth plant were used at different adsorbent metal ions ratios, and investigated the influence of pH, content time, metal concentration and

absorbent loading weight on the removal process (Abia et al., 2002; Singh et al., 2005; Abdel-Ghani and El-chaghaby, 2007).

Many researchers have studied chemical and biological pretreatments for enhancing conversion of lignocellulosic materials to protein enriched product (Vijaya et al 2007) . *Pleurotus pulmonarius* via solid state fermentation technique, through enhancement of *in vitro* digestibility and protein content an expense of crude fiber and fiber fraction contents .

The main objective of the current study is concerned with nutritional value upgrading of rice straw, banana leaves and water hyacinth leaves to protein enriched by-product via using combined mixed of them and white rot fungi (*Pleurotus ostreatus*) in solid state fermentation process

MATERIALS AND METHODS

MATERIALS

Rice straw and banana leaves were obtained from the experimental farm of Agricultural Research Center, Giza, Egypt. They were rinsed with water, air dried, then cut into about 1-2 cm. The chopped rice straw and banana leaves were dried at 70 °C for 24 hour.

White rot fungi , namely *Pleurotus ostreatus* NRRL-2366 was obtained from the National Center of Agricultural Utilization Research Service, US., Department of Agricultural, Peoria, Illinois, USA. The strain was maintained on potato dextrose agar medium (Difco Manual, 1979), then store at 4°C.

MICROBIOLOGICAL METHODS

PREPARATION OF GRAINS SPAWN

To prepare grains master spawn, wheat seeds or sorghum seeds were used.

Seeds were cleaned from debris, then soaked in water for overnight. Dead seeds were removed, then boiled in water 15 min. After cooling, the seeds were transferred to around bottle by occupying 2/3 of its volume and mixed with calcium carbonate 2% (w/w) and calcium sulphate 1% (w/w). Bottles were then sterilized, for 1hr at 121°C. After cooling, the sterilized bottles were inoculated with mycelial discs (5 mm diameter) which were born from the margins of 6 days old culture of *Pleurotus ostreatus*. The inoculated bottles were incubated at 25 °C for 15-20 days. The grains master spawn was used to inoculate bags containing (100 g) pasteurized maize stalks.

Solid state cultivation technique: The nutritional upgrading trial of rice straw or banana leaves were carried out in plastic bags containig 100 g of rice straw or banana leaves (straw or leaves of 1-2 cm length were pasteurized in hot water 90 °C for 2 hrs.). The moisture content of rice straw or banana leaves were adjusted to 70%. The bags were inoculated with 10-12g *Pleurotus ostreatus* spawn, in combined mixture with water hyacinth leaves(5,10,15,20,25 and 30g/ 100 g rice straw or banana

leaves) for each bag. The inoculated bags were incubated on 28 °C for 28 days (Darwish, 2000).

Preparation of water hyacinth binary mixtures: After chopped rice straw or banana leaves different combinations with water hyacinth leaves with different ratios (5, 10, 15, 20, 25 and 30 gm of water hyacinth leaves/100 gm rice straw or banana leaves). These combinations were made to explore their effect on nutritive value and digestibility.

Chemical analytical methods

Moisture content, ash, crude fiber, crude protein ether extract, neutral and acid detergent fiber were determined according to separate methods described in AOAC (2002). Total hydrolyzable carbohydrates were determined according to Montgomery (1961). Lignocellulosic fractions based on dry matter basis were determined according to the method of Van Soest and Robertson (1980).

In vitro disappearance

The *in vitro* dry matter disappearance (DMD) and organic matter disappearance (OMD) of samples were determined according to the two stages technique described by Tilley and Terry (1963). The rumen liquor was collected from fistulated sheep fed ration consisted of 70% wheat straw, 15% alfalfa hay and 15% concentrate feed mixture.

Statistical analysis

Statistical analysis for each separate collected data was done according to Gomez and Gomez (1984). The treatment means were compared using the least significant difference test (LSD) at the 5% level of probability as outlined by Waller and Duncan (1969).

RESULTES AND DISCUSSION

The chemical composition of these materials are given in Table 1&2 showing the limiting factors for their utilization by ruminants are low crude protein, high fiber and low available energy contents.

Data in Tables (1&2) show the biological treatment of rice straw and banana leaves by using different amounts of *Pleurotus ostreatus* inoculum, in solid state cultivation techniques. The obtained results revealed a significant increase ($p < 0.05$) of crude protein content in all treated samples, compared to control. The highest protein contents being 7.90% and 16.02% were obtained when rice straw and banana leaves were fermented using *Pleurotus ostreatus* for 28 days, respectively. Commonly, the protein content was increased in all treatments including both *Pleurotus ostreatus* compared treatment of rice straw and banana leaves without *Pleurotus ostreatus*.

The fiber fractions content decreased in all treatments with most significant decrease noted for all treatments compared to control

The obtained results also showed a decrease in cellulose and hemicelluloses contents compared to control. Gradual decrease in lignin content was noticed during different incubation periods. The extent of lignin degradation could be attributed to the ability of *Pleurotus ostreatus* to produce lignin degraded enzyme such as lignin peroxidase and manganese peroxidase which aid enzymatic degradation (Nerude and Misarcova,1995).

The review of the results indicates that rice straw and rice hulls are deficient in protein, energy, and minerals and their nutritive values are quite low. Physical, chemical and microbiological methods have been investigated to improve the digestibility and nutritive value of these by-products. Supplementation with energy, protein, minerals and vitamins resulted in improving the utilisation of the roughages.

In vitro dry matter disappearance (IVDMD) and *in vitro* organic matter disappearance (IVOMD) revealed their maximum values for treatment rice straw being 26.74 % and 50.20 %, respectively, compared to control being 29.68% and 56.60%, when added 10g inoculum of *Pleurotus ostreatus* ,but being 29.55% and 50.20%, respectively, compared to control being 31.60% and 56.20%, in banana leaves when added 12g inoculum of *Pleurotus ostreatus*

Deleted:

These obtained results came in agreement with those reported of by Mukherjee and Nandi (2004), Vijaya et al (2006) and Guides et al (2008), as they found that lignin decomposition (delignification) by fungus consequently leads to an increase in (IVDMD) which is considered a positive change in increasing the overall digestibility of feed composition. Zadrazil and Kamra (1989) reported improvement of *in vitro* digestibility after fermentation with *Pleurotus spp.*

In addition, these residues are needed to increase protenuaus compounds. This native composition of the crop residues make them unsuitable for animal feeding. Therefore, water hyacinth leaves should be take place, by mixing with rice straw and banana leaves with different ratios (5, 10, 15, 20, 25and 30g/100g substrate), it could be cleared that water hyacinth leaves contain high amounts of crude protein and minerals, in this study we made treatments combination of rice straw, banana leaves and water hyacinth leaves to upgrade the nutritive value and *in vitro* digestibility to possible can be used as fodder ruminants. Improving the digestibility and nutritive value of rice straw, banana leaves by using combination mixture technique, was accomplished. The structural chemical changes in the components of the resulted product were evaluated (Tables 3 &4).

The obtained results revealed increasing of crude protein in all treatments in comparison with control. High protein content being10.00% and 17.10% when mix 30g of water hyacinth leaves with 100 g rice straw or banana leaves, respectively. Commonly, the protein content increased with all treatments. Similar results were reported by Abd El-Rahman (1996), El-Shaer et al., (2005) and Abd El-Hamid et al., (2006).

The obtained results also revealed that crude fiber decreased with all treatments. Decrease in crude fiber values as responsibility of mixing was reported by Rogosic et al., (2005) and Boghuhn et al., (2006).

The obtained results also confirmed a decrease in fiber fractions contents. Cellulose and hemicellulose were also decreased with all tested treatments, whereas they decrease from 33.30% and 27.50% to 21.74% and 11.71%, respectively, when mixed 30 g of water hyacinth leaves with 100 g of rice straw, however, they being 16.88% and 8.97%, respectively, when mixed 30 g of water hyacinth leaves with 100 g of banana leaves. It was interested to determine gradual decrease in lignin in course of different ration. Degradation of lignin resulted in increasing of *in vitro* digestibility of rice straw, banana leaves and water hyacinth leaves mixture and confirming their uses as feed for ruminant animals. These results are in agree with the findings of **Blummel and Becker (1997)**; **Allam et al., (2006)** and **Patra (2007)**.

In vitro dry organic matter disappearance (IVOMD) showed high significant ($P<0.05$) increase. In general, the obtained results cleared these treatment succeeded to improve the nutritive value of rice straw, banana leaves and water hyacinth leaves mixture to be used for ruminant feeding. These results are agreement with **Khattab et al., (1999)**; **Patra et al., (2006)**; **Darwish and Ali (2005)**; **Agarwal et al., (2006)**; **Chumpawadee et al., (2007)**; **Sallam et al., (2007)** and **Darwish and Bakr (2010)**-

CONCLUSION

This study indicated that treatments of rice straw or banana leaves as agricultural by product with combination water hyacinth leaves and *Pleurotus ostreatus* can increase crude protein and improve digestion. Furthermore, this study opened useful applied access for production of rice straw or banana leaves based products of good quality. Moreover, the current study can add applied values based on fungal treatment of non beneficial agricultural waste.

Table (1) : Biological treatment of rice straw using different amounts of *Pleurotus ostreatus* inoculum in solid state fermentation technique (incubated at 28° C for 4- weeks)

| Fungal inoculum amounts (g) | Biomass recovery (g) | Crude protein obtained | | Net gained protein (g) | Fungal mycelial dry wt. (g/100g) | Undegraded substrate (g) | Degraded substrate (g) | Bioconversion efficiency (g) | In vitro disappearance | | Chemical analysis of biomass recovery | | | |
|-----------------------------|----------------------|------------------------|------|------------------------|----------------------------------|--------------------------|------------------------|------------------------------|------------------------|-------|---------------------------------------|-------------|---------|-------|
| | | % | (g) | | | | | | DMD | OMD | Hemicellulose % | Cellulose % | Lignin% | Ash % |
| 2 gm | 74.24 | 2.80 | 2.10 | 0.50 | 1.43 | 72.81 | 27.19 | 1.84 | 40.20 | 41.80 | 28.10 | 35.10 | 9.90 | 16.80 |
| 4 gm | 75.30 | 4.70 | 3.54 | 2.40 | 6.86 | 68.44 | 31.56 | 7.61 | 42.60 | 45.50 | 22.90 | 35.20 | 9.30 | 17.10 |
| 6 gm | 76.24 | 4.90 | 3.74 | 2.60 | 7.43 | 68.81 | 31.19 | 8.34 | 46.10 | 48.20 | 23.30 | 33.80 | 9.10 | 17.30 |
| 8 gm | 78.20 | 6.30 | 4.93 | 4.00 | 11.43 | 66.77 | 33.23 | 12.04 | 46.30 | 48.20 | 22.70 | 33.00 | 8.10 | 17.50 |
| 10 gm | 76.00 | 7.90 | 6.00 | 5.60 | 16.00 | 60.00 | 40.00 | 14.00 | 50.20 | 56.60 | 20.20 | 31.30 | 8.30 | 18.00 |
| 12 gm | 77.02 | 7.80 | 6.01 | 5.50 | 15.71 | 61.31 | 38.69 | 14.22 | 55.55 | 59.10 | 19.00 | 31.00 | 8.85 | 18.10 |
| **Control | 100.00 | 2.30 | 2.30 | — | — | 100.00 | — | — | 26.74 | 29.68 | 27.50 | 33.30 | 11.60 | 16.70 |

Table 1 (Continued; Foot notes)

a, b ,c Means in the same column with different superscripts significantly different (p<0.05)

IVDMM *in vitro* dry matter disappearance

IVOMD *in vitro* organic matter disappearance *Crude protein content of *Pleurotus ostreatus* (35%)

Table (3) : Biological treatment of rice straw and water hyacinth leaves mixture by using **Pleurotus ostreatus* in solid state fermentation (incubated at 28 ° C for 4- Weeks)

| Fermented media mixture ratio | Dry matter% | Total hydrolysable carbohydrates | Ether extract | Crude protein | Crude Fiber | <i>In vitro</i> disappearance | | Chemical analysis of biomass recovery | | | |
|--------------------------------------|-------------|----------------------------------|---------------|---------------|-------------|-------------------------------|-------|---------------------------------------|------------|----------|-------|
| | | | | | | DMD | OMD | Hemi-cellulose % | Cellulose% | Lignin % | Ash % |
| *Rice straw(RS) (control)untreated | 86.60 | 34.10 | 1.20 | 2.30 | 39.40 | 26.74 | 29.68 | 27.50 | 33.30 | 11.60 | 16.70 |
| **Water hyacinth leaves(Wh)untreated | 85.87 | 32.50 | 3.05 | 20.90 | 20.80 | 40.44 | 52.81 | 13.64 | 15.15 | 5.73 | 17.99 |
| RS+W h (5%) untreated | 85.80 | 25.25 | 1.30 | 3.23 | 38.47 | 42.50 | 46.70 | 18.30 | 35.35 | 13.00 | 17.55 |
| RS+W h (5%) treated | 78.50 | 20.01 | 1.54 | 7.40 | 33.86 | 44.30 | 53.20 | 19.32 | 34.72 | 12.27 | 17.60 |
| RS+W h (10%) untreated | 83.90 | 22.95 | 1.40 | 5.70 | 37.54 | 42.80 | 48.80 | 20.29 | 33.72 | 11.90 | 17.00 |
| RS+W h (10%) treated | 77.16 | 20.72 | 1.47 | 7.50 | 33.12 | 46.30 | 54.80 | 16.47 | 34.89 | 9.99 | 16.35 |
| RS+W h (15%) untreated | 83.50 | 23.88 | 1.59 | 6.30 | 36.61 | 42.80 | 46.80 | 17.24 | 31.52 | 7.51 | 16.02 |
| RS+W h (15%) treated | 75.51 | 20.00 | 1.63 | 7.60 | 33.07 | 46.50 | 54.70 | 14.18 | 28.39 | 7.05 | 16.23 |
| RS+W h (20%) untreated | 82.48 | 23.80 | 1.56 | 6.50 | 35.68 | 42.90 | 47.80 | 14.11 | 25.72 | 6.33 | 15.95 |
| RS+W h (20%) treated | 72.00 | 16.59 | 1.64 | 7.90 | 31.88 | 47.40 | 55.10 | 13.56 | 23.19 | 5.58 | 15.99 |

| | | | | | | | | | | | | | |
|-------------------|---|-------|-------|-------|------|-------|-------|-------|-------|-------|-------|------|-------|
| RS+W untreated | h | (25%) | 81.73 | 23.70 | 1.67 | 6.83 | 34.75 | 43.50 | 48.10 | 14.02 | 21.88 | 5.83 | 16.11 |
| RS+W treated | h | (25%) | 71.20 | 18.23 | 1.94 | 8.00 | 28.48 | 48.50 | 55.10 | 14.26 | 21.39 | 5.55 | 16.20 |
| RS+W untreated | h | (30%) | 81.80 | 23.00 | 1.53 | 8.20 | 33.82 | 43.60 | 48.60 | 14.11 | 22.00 | 6.10 | 16.43 |
| RS+W treated | h | (30%) | 70.00 | 17.21 | 1.69 | 10.00 | 25.37 | 49.50 | 55.70 | 11.71 | 21.74 | 5.59 | 16.73 |

Table 3 (Continued; Foot notes)

a, b ,c Means in the same column with different superscripts significantly different ($p < 0.05$)

* RS Rice Straw without any treatment

**W h Water hyacinth leaves without any treatment

IVDMM *in vitro* dry matter disappearance

IVOMD *in vitro* organic matter disappearance

Table (2) : Biological treatment of banana leaves using different amounts of *Pleurotus ostreatus* inoculums

in solid state fermentation technique (incubated at 28° C for 4- weeks)

Table 2 (Continued; Foot notes)

| Fungal inoculum amounts (g) | Biomass recovery (g) | Crude protein obtained | | Net gained protein (g) | Fungal mycelial dry wt. (g/100 g) | Undegraded substrate (g) | Degraded substrate (g) | Bioconversion efficiency (g) | In vitro disappearance | | Chemical analysis of biomass recovery | | | |
|-----------------------------|----------------------|------------------------|-------|------------------------|-----------------------------------|--------------------------|------------------------|------------------------------|------------------------|-------|---------------------------------------|-------------|----------|-------|
| | | % | (g) | | | | | | DM D | OM D | Hemi-cellulose % | Cellulose % | Lignin % | Ash % |
| 2 gm | 78.78 | 14.00 | 11.03 | 0.40 | 1.14 | 77.64 | 22.36 | 1.80 | 47.80 | 54.20 | 19.87 | 28.70 | 8.34 | 12.00 |
| 4 gm | 79.35 | 14.44 | 11.46 | 0.84 | 2.40 | 76.95 | 23.05 | 3.64 | 48.70 | 55.30 | 19.45 | 27.75 | 8.00 | 12.25 |
| 6 gm | 79.10 | 14.78 | 11.70 | 1.18 | 3.37 | 75.73 | 24.27 | 4.86 | 49.00 | 55.20 | 17.42 | 25.84 | 7.17 | 12.60 |
| 8 gm | 80.00 | 15.01 | 12.01 | 1.41 | 4.03 | 75.97 | 24.03 | 5.87 | 49.50 | 55.70 | 14.13 | 24.44 | 6.30 | 12.88 |
| 10 gm | 79.23 | 15.17 | 12.11 | 1.57 | 4.49 | 74.74 | 25.26 | 6.22 | 49.20 | 56.10 | 13.10 | 23.86 | 5.63 | 12.95 |
| 12 gm | 80.04 | 16.04 | 12.84 | 2.44 | 6.97 | 73.07 | 26.93 | 9.10 | 50.20 | 56.20 | 12.80 | 27.67 | 5.49 | 13.05 |
| **Control | 100.00 | 12.50 | 12.50 | — | — | 100.00 | — | — | 29.55 | 31.60 | 16.25 | 26.30 | 8.50 | 11.96 |

a, b ,c Means in the same column with different superscripts significantly different (p<0.05)

IVDMM *in vitro* dry matter disappearance IVOMD *in vitro* organic matter disappearance

***Control treatment (untreated banana leaves) Crude protein content of *Pleurotus ostreatus* (35%)

Table (4) : Biological treatment of banana leaves and water hyacinth leaves mixture by using *Pleurotus ostreatus in solid state fermentation (incubated at 28 ° C for 4- Weeks)

| Fermented media mixture ratio | Dry matter% | Total hydrolysable carbohydrates | Ether extract | Crude protein | Crude Fiber | In vitro disappearance | | Chemical analysis of biomass recovery | | | |
|--------------------------------------|-------------|----------------------------------|---------------|---------------|-------------|------------------------|-------|---------------------------------------|------------|---------|-------|
| | | | | | | DMD | OMD | Hemi-cellulose % | Cellulose% | Lignin% | Ash % |
| *Banana leaves(BI)(control)untreated | 88.40 | 38.55 | 1.75 | 12.50 | 28.50 | 29.55 | 31.60 | 16.25 | 26.30 | 8.50 | 11.96 |
| **Water hyacinth leaves(Wh)untreated | 85.87 | 32.50 | 3.05 | 20.90 | 20.80 | 40.44 | 52.81 | 13.64 | 15.15 | 5.73 | 17.99 |
| BI+W h (5%) untreated | 85.98 | 30.55 | 2.02 | 12.93 | 28.22 | 49.10 | 54.60 | 15.68 | 24.46 | 9.60 | 12.26 |
| BI+W h (5%) treated | 78.20 | 23.99 | 2.20 | 14.10 | 25.49 | 52.60 | 58.20 | 14.38 | 22.96 | 9.10 | 13.42 |
| BI+W h (10%) untreated | 86.59 | 29.50 | 2.32 | 13.34 | 27.90 | 49.50 | 55.70 | 15.54 | 23.47 | 9.46 | 13.60 |
| BI+W h (10%) treated | 77.14 | 21.00 | 2.58 | 15.40 | 24.44 | 54.60 | 59.60 | 14.52 | 22.65 | 8.99 | 13.95 |
| BI+W h (15%) untreated | 81.04 | 23.10 | 2.40 | 13.77 | 27.60 | 55.60 | 59.80 | 14.02 | 22.57 | 8.18 | 14.20 |
| BI+W h (15%) treated | 76.00 | 19.05 | 2.95 | 15.80 | 23.81 | 56.20 | 60.20 | 12.41 | 20.10 | 7.39 | 14.47 |
| BI+W h (20%) untreated | 84.83 | 25.90 | 2.73 | 14.18 | 27.30 | 53.80 | 59.60 | 11.80 | 21.00 | 7.37 | 14.70 |
| BI+W h (20%) treated | 75.50 | 18.00 | 3.30 | 16.70 | 22.92 | 54.20 | 60.10 | 11.25 | 19.35 | 6.30 | 15.30 |
| BI+W h (25%) untreated | 81.43 | 22.22 | 2.77 | 14.60 | 27.00 | 54.20 | 60.60 | 10.90 | 19.00 | 6.00 | 14.95 |

| | | | | | | | | | | | |
|------------------------|-------|-------|------|-------|-------|-------|-------|-------|-------|------|-------|
| Bl+W h (25%) treated | 73.85 | 16.10 | 3.45 | 17.00 | 21.90 | 60.60 | 54.20 | 10.55 | 17.85 | 5.85 | 15.90 |
| Bl+W h (30%) untreated | 79.51 | 19.65 | 2.98 | 15.02 | 26.60 | 54.70 | 56.77 | 10.34 | 17.15 | 5.75 | 15.29 |
| Bl+W h (30%) treated | 71.24 | 14.44 | 3.84 | 17.10 | 19.78 | 61.90 | 54.60 | 8.97 | 16.88 | 5.35 | 16.10 |

Table 4 (Continued; Foot notes)

a, b ,c Means in the same column with different superscripts significantly different ($p < 0.05$)

* Bl Banana leaves without any treatment

**W h Water hyacinth leaves

IVDMM *in vitro* dry matter disappearance IVOMD *in vitro* organic matter disappearance

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