

# DOMESTIC SEWAGE WASTEWATER IRRIGATION ON QUALITATIVE EVALUATION OF NAPIER GRASS (*PENNISETUM PURPUREUM*) AND ITS IMPACT ON COW MILK PARAMETERS REFERENCE TO ENVIRONMENTALLY SUSTAINABLE FOOD SECURITY

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## ABSTRACT

Domestic sewage wastewater irrigation for the agricultural fields is highly promoted for sustainable water resource management. The present case study investigated domestic sewage wastewater irrigation influences on the agro botanical parameters and biomass productivity of Napier grass. Further, the quality of milk (proximate and mineral composition) has also been checked from cows fed with wastewater-irrigated Napier grass. The study has been conducted in 5 different agricultural fields (4 wastewater irrigated fields and 1 well-water irrigated field as control (A-E)). The results of the study reveal that the physiochemical parameters of all the domestic wastewater samples were within the permissible limit of (FAO-1985)/(WHO-1993) and (BIS-1986) standards. However, all the parameters were found to be higher when compared with the control sample. Interestingly, the agro botanical characteristics of Napier grass grown in semi-urban agro field (D) irrigated with wastewater were found to be significantly ( $P < 0.05$ ) higher. However, when compared to all experimental samples, significantly ( $P < 0.05$ ) higher values were observed for proximate compositions of both Napier grass and cow milk samples from agro field A (semi-urban). Moreover, macro-micronutrients and heavy metal profiles in the plant and milk samples were also found to be within acceptable/permissible limits. At this juncture, the results of the study reveal that, due to different mitigation measures, domestic sewage wastewater can be used as a potential organic liquid fertilizer, which will eventually reduce the extraction of freshwater and improve the wastewater reuse for the cultivation of such fodder crops and thereby ensure nature-based and sustainable water resource management.

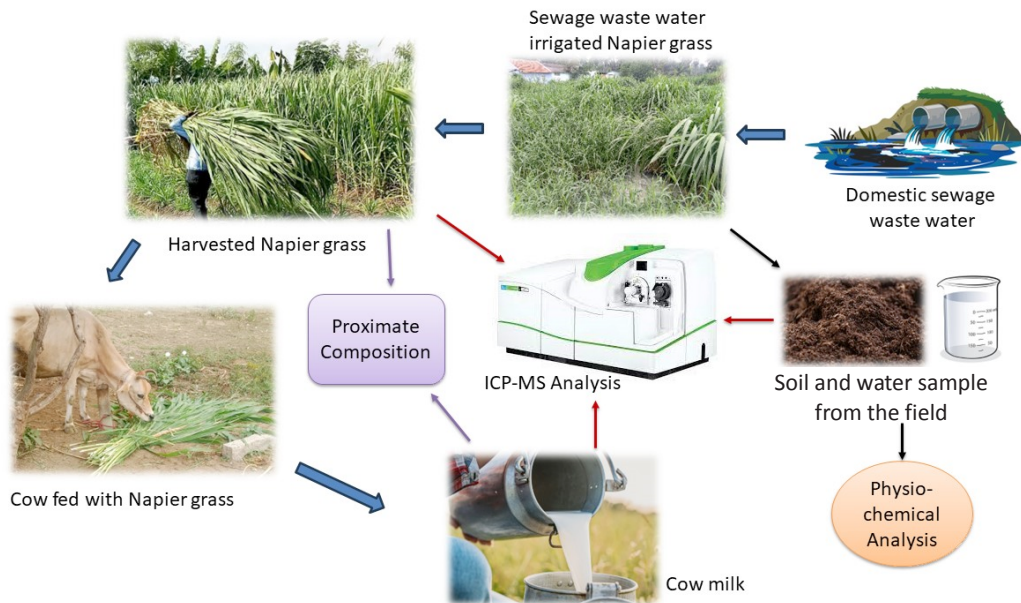
**KEY WORDS:** Domestic wastewater irrigation, Napier grass, Proximate composition, Mineral profiles, Cow milk components.

## INTRODUCTION

Water is one of the elements required for life to exist on earth. Water is a necessity in domestic, agricultural, industrial, and other related operations (Adejumo and Adebisi, 2020). Global issues including rising water usage, pollution, and climate change are to blame for water scarcity. Water scarcity is a threat to 80% of the world's population (Pulla *et al.*, 2018). Crop cultivation via wastewater irrigation is referred to as wastewater agriculture. In

this context, urban wastewater is used as a source of irrigation for farmers in urban and semi-urban regions in many cities in developing countries (Miller-Robbie *et al.*, 2017). In locations with a shortage of freshwater resources, using wastewater for agriculture is a crucial management method with potential environmental and economic advantages. Water reuse can provide alternatives to existing water supplies and be used to enhance water security, sustainability, and climate resilience. The approach has numerous advantages, including

## GRAPHICAL ABSTRACT



nutrient recycling, water conservation, and the reduction on over exploitation of surface and groundwater resources (Jaramillo *et al.*, 2017).

Reusing wastewater for agricultural irrigation decreases the need to withdraw water from water sources (Ramirez *et al.*, 2021). It is a viable way to lower the demand for freshwater while maintaining zero water discharge and preventing pollution load in receiving sources. In the modern day, it is necessary to consider the infrastructure for disposing of urban waste, wastewater agriculture techniques, the quality of the water used, the effects on human health, and the degree of institutional awareness of wastewater-related concerns (Schellenberg *et al.*, 2020). Particularly, large volumes of residential wastewater are produced in cities as a result of the ever-growing population. The uncontrolled dumping of this wastewater contributes to the contamination of the groundwater, air, and soil. In a developing countries like India, the cost of sewage wastewater treatment for recycling is too high to be regular practise. However, because most of the nutrient load is exerted by this wastewater, it might be used as irrigation water for some trees, plants, and crops, which could result in an increase in agricultural output and plantations. It may be able to provide both micro and macro nutrients (NPK) and carbon nutrients as liquid fertilizers to boost crop or plant growth (Singh *et al.*, 2012).

Numerous uses for sewage water irrigation exist,

including irrigation of crops, aquaculture, landscapes, and fictitious groundwater recharging (Khan, 2018). Most places in the world consider this to be one of the oldest and most well-known customs and nature based traditional method. Around 20 million hectares of land worldwide are thought to be used for wastewater irrigation. The majority of crop plants, including lettuce, mangoes, tomatoes, and coconuts have been found to be watered with sewage water, much of which is untreated (Thebo *et al.*, 2017).

A key part of the Indian economy is livestock. The livelihood of almost 20.5 million people depends on livestock. Compared to an average of 14% for all rural households, livestock contributed 16% of the revenue for small farm households. For two-thirds of rural communities, livestock is the main source of income. Additionally, it employs around 8.8% of India's population. India has a tremendous supply of livestock. The livestock industry makes up 25.6% of all of agriculture's GDP and adds 4.11% to it (Das *et al.*, 2020). Because of the expanding population and better living conditions, there is currently a growing demand for ruminant animal products. However, there are significant production barriers in the smallholder livestock industry due to feed shortages and poor fodder quality during the dry season (Godde *et al.*, 2021). Smallholder farmers are looking for inexpensive alternatives because of the pressure that climate change, economic considerations, and anthropogenic factors are

placing on conventional feeds to remain unaffordable. The least expensive types of animal feed are still pasture and fodder (Belay *et al.*, 2017). The optimum pasture and fodder species should be highly productive, tasty, and contain sufficient amounts of nutrients that are readily digested to meet the needs of livestock (Filleau *et al.*, 2018).

Under recent climate change scenario of continuous drought, water contamination and water scarcity, the availability of high-quality feed and forage supplies is crucial for sustainable livestock production (Varijakshapanicker *et al.*, 2019). The increasing severity of the livestock feeding issue has made dairy and fattening farms most competitive and difficult to maintain under an open market trading policy. Farm-level production of fodder is quite rare, although in milk pocket regions, farmers typically grow fodder for their animals as well as for sale in the market. By offering the most affordable form of feed for livestock, fodder crops play a crucial role in the agricultural economies of developing nations. Milk is essential for the physical and mental development of humans. Dairy animals function best when there is a steady and sustainable supply of good fodder in sufficient quantities. Therefore, the lack of enough quality feed is the major obstacle to profitable animal production in Least Developed Countries (Erickson *et al.*, 2020).

*Pennisetum purpureum* (synonym *Cenchrus purpureus*) is an important perennial forage grass that is closely related to pearl millet (*P. americanum*). It is a rhizomatous big grass that can be found on degraded soils, pastures, streams, floodplains, marshes, swamps, riverbanks, and agricultural fields (Pandey *et al.*, 2020). More than 80% of the forages fed to dairy cattle kept in stalls are derived from napier grass fodder. It is frequently employed in cut-and-carry feeding systems and is becoming more significant in other agricultural systems. Napier grass is a preferred fodder due to its many advantageous qualities, which include high yield per unit area, resistance to periodic drought and flood conditions and highly suitable for irrigation with domestic sewage waste water. Since long back, napier grass has been cultivated under domestic sewage wastewater irrigation in order to produce biomass production including enrichment of nutrients bio-availability and water recycling. Further, napier grass is a suitable feedstock for the production of cellulosic ethanol due to its high biomass yield, which also offers the added benefits of enhancing water quality and supplying

sustainable energy (Iwai *et al.*, 2015). It can withstand against repeated cutting and regenerates quickly, giving forth tasty green shoots. Therefore, improving knowledge-based utilisation and conservation of the existing Napier grass resources promises to have a significant positive impact on cattle production value chains (Negawo *et al.*, 2017).

The physical characteristics of soil, such as bulk density, water retention, and hydraulic conductivity, are improved by sewage irrigation. Chemical properties such as the presence of a high amount of organic carbon and the build-up of soil available N, P, K, and micronutrient status in the sewage-irrigated soils improve the soil fertility status to some extent, while physicochemical properties such as pH and electrical conductivity were increased due to sewage water having the high amount of salts and degrading the soil structure to some extent (Gurjar *et al.*, 2017).

In agriculture and animal production value chains, where in-depth knowledge of the available resources is necessary, genetic resources play a crucial role. The main justifications for the protection and utilisation of the genetic resources that are currently available are thought to be accurate passport, characterization, and assessment data, together with a general comprehension of the diversity of the genetic resources (Negawo *et al.*, 2017). There are currently more than 300 Napier grass germplasm preserved in various gene banks with the collaboration of United Nations Organization and the FAO.

The purpose of the present work was to study the status of plant growth and proximate composition with wastewater irrigation in urban and semi-urban areas of Coimbatore city, Tamil Nadu, India, with respect to growth and yield as compared to that of groundwater irrigated crops. The effective handling of wastewater and soil pollution together will benefit not only the environment and the ecosystem, but also improve the quality of human life. The best way to do this is the cultivation of Napier grass with wastewater leads to reduced wastewater management and feed for the livestock which is economically beneficial to mankind. Because of its wide range of flexibility and high protein content, dairy cows produce more milk, which benefits many farmers particularly small scale farmers of the peri-urban agro-ecosystem. In this context, the present research study has been carried out in view of effect of domestic sewage wastewater irrigation on qualitative evaluation of napier grass and it fed with

cow for milk quality assessment.

## METHODS

### Study Area

One of the largest metropolises in the Indian state of Tamil Nadu is Coimbatore, often known as Kovai or occasionally spelt Covai. The Western Ghats encircle it, and it is situated on the Noyyal River's banks. Coimbatore is the 16<sup>th</sup> largest urban agglomeration in India and the second largest city in Tamil Nadu after Chennai. The study areas were shown in Fig.1. In the Coimbatore district used for this investigation, samples were taken from two semi-urban areas (Nanjundapuram-A and Vellalore-C), two urban areas (Ukkadam-B and Singanallur-D) which are all irrigated with domestic wastewater and one rural region (Narasipuram -E control).

### Sample collection

**Plant sample:** Napier grass was harvested about 10–15 cm above the ground from all the study areas within the interval time period of 6-7 weeks (42-49 days) from the previous cuttings (the recommended period for feeding the Napier grass for animals). Samples were powdered to pass through a 2 mm sieve after being oven dried at 60 °C to a constant weight.

**Milk:** Cow milk samples which fed with respective Napier grass were collected from the study area and it is stored in deep freeze (-20 °C) for further analysis.

**Water:** In 100-ml polythene bottles that had already been cleaned, independent water samples were collected from the study area. Water samples were stored in an insulated field kit with ice before being delivered to the lab. The parameters of common water were examined based on the calibration procedures employed by the American Public Health Association for the evaluation of water and wastewater (APHA 2017). A number of characteristics were examined, including pH, electrical conductivity (EC), suspended solids (SS), total dissolved solids (TDS), BOD, DO, COD, total hardness, magnesium hardness, calcium hardness, nitrate, sulphate, and phosphorus (P).

**Soil:** Using a soil hand auger, soil samples were collected from the topsoil and the (0-20 cm) depth. Soil samples of 1 kg were collected in triplicate (n=3) from each sampling site using the quadrat method. The plastic bags, which were 3 mm thick, were used to collect the samples and were mixed thoroughly, air dried and passed through a 2mm mesh sieve.

### Proximate analysis

Using the Moisture Analyzer MA35 (Sartorius AG, Germany) at 105 °C, the moisture content of the samples was ascertained. The techniques described in the Association of Official Analytical Chemists were also used to determine the crude lipid (Soxhlet extraction), crude fibre, and ash concentrations. The total nitrogen was calculated using the Micro-Kjeldahl method, and crude protein ( $N \times 6.25$ ) was calculated using a nitrogen protein conversion factor. The difference was used to calculate the crude

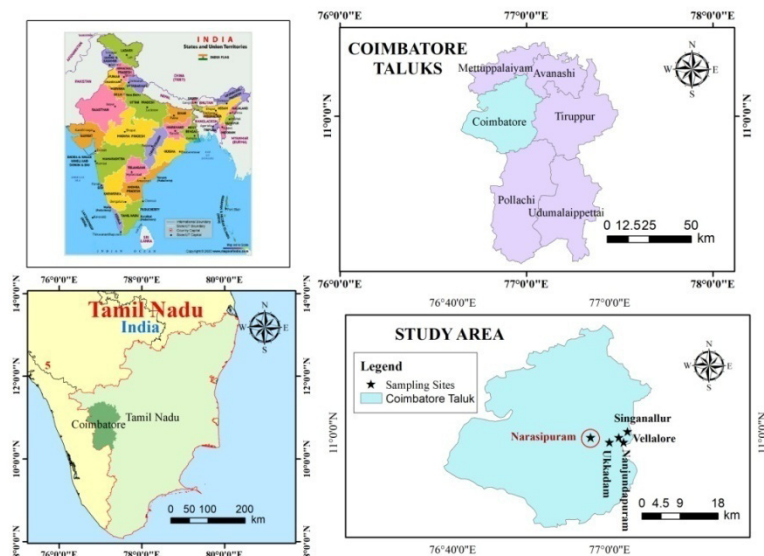


Fig. 1. Study Area

carbohydrate content, commonly known as nitrogen-free extractives (NFE). In terms of g/100 g DM, the approximate composition was stated. By multiplying the percentages of crude protein, crude lipid, and NFE by 16.7, 37.7, and 16.7, respectively and the gross energy (KJ) was calculated according the method of Association of Official Analytical Chemists (AOAC, 2019).

### Mineral analysis

Triacid was used to digest the samples.  $\text{HNO}_3$ ,  $\text{H}_2\text{SO}_4$  and  $\text{HClO}_4$  were combined in the triacid mixture in the proportions of 9:2:1; 200 mg of sample was combined with 10 ml of triacid and digested at 80 °C. Following digestion, the samples were diluted to 100 ml and the minerals and heavy metals were analysed through the ICP-MS -NexION 300X, Perkin Elmer, USA (Wilschefska *et al.*, 2019).

### Statistical analysis

The data were subjected to a one-way analysis of variance (ANOVA), and the significance of difference between means was determined by Duncan's multiple range test ( $P < 0.05$ ) using SPSS (version 21, SPSS Inc., Waker drive Chicago, USA). Values expressed are means of triplicate determination  $\pm$  standard deviation.

## RESULTS AND DISCUSSION

### Physico-chemical parameters of sewage wastewater irrigation sample

The physico-chemical parameters are very essential and important to check the water, before it is used for drinking, domestic or agricultural purposes. The result of physico chemical properties of the domestic sewage wastewater and well water is represented in Table 1, and this was compared with the FAO/WHO/BIS standard for irrigation. pH is a measure of the concentration of hydrogen ions ( $\text{H}^+$ ) in water. All the samples were reported with optimum pH for irrigation based on the standards. The electrical conductivity is an important parameter used to estimate the level of dissolved salts in water. The electrical conductivity was reported highest in sample A and lowest in control but all samples are within the permissible limits. On the other hand, the total dissolved solids" refer to any minerals, salts, metals, cations or anions dissolved in water. The parameters such as TDS, TSS, TS, Total Hardness, Sulphate, Nitrate, Phosphate, Chloride, Sodium and Potassium in all the wastewater samples are found to be comparatively higher than the control and this might be due to the combination of inorganic salts, principally calcium, magnesium, potassium,

**Table 1.** Physico-chemical parameters of sewage wastewater irrigation samples

Parameters	Sample-A	Sample-B	Sample-C	Sample-D	Sample-E (Control)	FAO-1985/ WHO-1993	Indian Standards (BIS)-1986
pH	7.68	7.14	7.32	7.14	7.02	6.5-8.5	6.5-8.5
Conductivity (mS $\text{cm}^{-1}$ )	2.32	2.18	1.59	1.64	1.35	3.0	2.25
TDS (mg $\text{l}^{-1}$ )	1506	1432	1064	1136	860	2000	2100
TSS (mg $\text{l}^{-1}$ )	336	320	302	356	246	-	-
TS (mg $\text{l}^{-1}$ )	1842	1752	1366	1492	1106	-	-
Total-Hardness(mg $\text{l}^{-1}$ )	455	380	320	365	305	-	300-600
Ca-Hardness (mg $\text{l}^{-1}$ )	235	255	210	225	190	250	-
Mg-Hardness (mg $\text{l}^{-1}$ )	120	125	110	130	115	-	-
Nitrate (mg $\text{l}^{-1}$ )	47	42	36	40	34	-	-
Phosphate (mg $\text{l}^{-1}$ )	0.78	1.02	1.24	1.08	0.76	2	-
Sulphate(mg $\text{l}^{-1}$ )	40	32	26	38	20	1000	1000
Chloride (mg $\text{l}^{-1}$ )	298	364	272	418	226	1100	600
Alkalinity (mg $\text{l}^{-1}$ )	278	252	294	304	212	-	200-600
DO (mg $\text{l}^{-1}$ )	5.68	4.26	3.72	4.80	1.68	6.0	-
BOD (mg $\text{l}^{-1}$ )	36.4	54.2	42.6	28.4	15.4	100	-
COD (mg $\text{l}^{-1}$ )	112.8	172.6	140.6	105.2	62.4	80-500	-
Na (mg $\text{l}^{-1}$ )	184	258	196	164	106	900	-
K (mg $\text{l}^{-1}$ )	15.8	13.2	18.4	14.6	10.4	-	-

TDS-Total dissolved solids, TS-Total solids, TSS- Total suspended solids, Ca-Calcium, Mg-Magnesium, DO-Dissolved Oxygen, BOD- Biological oxygen demand, COD- Chemical Oxygen demand, Na-Sodium, K- Potassium, FAO- Food and Agricultural Organization, WHO- World Health Organization, BIS- Bureau of Indian standards.

sodium, bicarbonates, chlorides, and sulphates and some small amounts of organic matter that are dissolved in water. Further, dissolved oxygen in irrigation water is playing a vital role in plant growth and health. When properly delivered to the root zone, high dissolved oxygen levels can drastically increase a plant's ability to utilize nutrients, defend it from pathogens, and increase crop yields. DO content of the studied water samples was at a level in which it is beneficial and safe for the plant's growth. Nonetheless, biological oxygen demand (BOD) of wastewater is an important criterion for judging the suitability of wastewaters for irrigation. Wastewaters with a wide range of BOD (0 to 1000 mg/l) were used for irrigation of a variety of crops. In the present investigation, all the samples were reported with optimum BOD for irrigation based on the standards. However, the physico-chemical parameters of the four wastewater samples were slightly higher than the control but all the above-mentioned parameters were found to be within the permissible limits which are also optimum for irrigation of crops. The similar results were also reported in the previous study (Dhanya *et al.*, 2020). However, the periodic monitoring of the physicochemical parameters of the domestic sewage wastewater before irrigation has to be done for environment and health safety measures.

#### **Agrobotanical characteristics of sewage wastewater irrigated Napier grass**

The effect of wastewater irrigation on the growth parameters of Napier grass has been given in Table 2 which include plant height, number of leaves, stem length, and dry biomass (Kg)/plant are all contributing factors to fodder yield in forage crop production (Patel *et al.*, 2018). The plant height of the samples ranges from 1.47 m to 2.19 m in which, the highest value was noted in sample D and lowest value was noted in sample C. The leaf length ranged from 1.02 m to 1.36 m. Similarly, other parameters such as number of leaves, fresh weight and dry matter of harvested plant were found to be highest in sample D and the lowest values were observed in sample C. Further, it is evident that the plant growth parameters which are higher or equal to the control sample might be due to nutrient enrichment such as micro-macro nutrients in the irrigated wastewater. A significant difference ( $P < 0.05$ ) between the plant height and number of leaves among the samples were also observed. However, as concern the terms

of leaf length and total dry matter there was no significant difference ( $P < 0.05$ ) noted between the samples C & E and A & B respectively. However, there is no significant difference ( $P < 0.05$ ) between the samples D & E, A & E and A, C, & B in the fresh weight of the samples. Further, the fresh matter value of the samples, D & E significantly ( $P < 0.05$ ) different from other samples such as B & C. However, sample E did not show any significant difference when compared to the sample A. In general, the agrobotanical characteristics of sewage wastewater irrigated napier grass has showed higher growth and yield performance and it was found to be similar to that of previous field study in the suburban area in the south western part of Mysore city, Karnataka, India (Alghobar *et al.*, 2016) and in Veterinary College and Research Institute, Orathanadu, Thanjavur, Tamil Nadu, India (Senthil Kumar *et al.*, 2021). Therefore, the present study reveals that the sewage wastewater irrigation is found to be more suitable for sustainable growth and biomass production of napier grass.

#### **Proximate composition of the sewage wastewater irrigated Napier grass**

The results of proximate composition are in the table 3. The proximate composition, which includes analyses of moisture, crude ash, crude protein, crude fat, and crude fibre, gives a general summary of the nutritional value of a feed. The nutritional value of Napier grass is crucial to this study because it affects the growth of ruminants. In order to effectively produce livestock, the need of establishing the nutritional value of grass forages in livestock nutrition is absolutely essential (Hapsari *et al.*, 2016).

For all of the study locations, the moisture content ranged from 6.93 to 8.67 % but the moisture content of the fresh weight samples ranges from 70.58 to 79.16 %. The moisture content, which serves as an important scientific measure, indicates how much water is present in the plants (Jin *et al.*, 2017). Napier grass would have a very limited shelf life, be subject to rapid deterioration, and require drying if preservation was sought, according to the moisture content of the plant. This plant may be able to help with quick rehydration in cases of animal dehydration, according to the moisture content. The lowest value was obtained in sample A (6.93 %) and the highest value was obtained in sample C (8.67%). There is no significant difference ( $P < 0.05$ ) between the samples B, C, D, & E and between the samples E&A.

Among the proximate composition of the plant samples, ash content poses high concentration of minerals and the ash content of the presently investigated Napier grass samples ranged between 11.51 and 13.75 dm%. Hence, the irrigation with sewage wastewater results in high mineral absorption and the increased ash level indicates that these samples are rich in minerals. On the other hand, ash content is typically considered to be a measure/bioindicator of mineral content in the food; as a result, ash analysis are crucial for evaluating nutrient labelling, material quality, microbial stability, and standard operating procedure in the food/feed industries (Md Noh *et al.*, 2020). In the present study, there is no significant difference on ash values ( $P < 0.05$ ) were found between the samples A (13.75%), B (12.51%), & E (13.1%) and between the samples C (11.71%) & D (11.51%). Further, the average ash content of the studied samples was found to be 12.51 % which is similar to that of previous studies (13%-14%) reported by several authors (Worqlul *et al.*, 2021; Lounglawan *et al.*, 2014; Zewedu *et al.*, 2002; Senthil Murugan *et al.*, 2016).

Muscle growth, milk protein outputs, illness resistance, the reproductive system, and bodily functioning maintenance are all impacted by protein, a nitrogen-derived substance that is essential for ruminant growth and development (Erickson *et al.*, 2020). The crude protein of napier grasses collected from urban, semi-urban (irrigated with sewage wastewater) and rural areas (irrigated with well water) ranged from 10.54 % to 12.60 %. Further, there was no significant difference ( $P < 0.05$ ) observed among the samples and the average value of crude protein content (11.63 %) was found to be similar to that of previous reports (Worqlul *et al.*, 2021; Lounglawan *et al.*, 2014; Mutimural *et al.*, 2018; Maleko *et al.*, 2019; Sarker *et al.*, 2018; Aganga *et al.*,

2005; Zewedu *et al.*, 2002; Butterworth *et al.*, 1965; Senthil Murugan *et al.*, 2016). The CP content of napier grass feed samples obtained was above the critical level ( $> 7$  % CP) which is necessary for voluntary ruminants' feed intake in sustaining the rumen microflora, optimal rumen digestibility, post rumen digestibility nutrient absorption and milk production (Beigh *et al.*, 2017).

The range of crude fat concentration in this study is 5.51% to 7.06%. Crude fat is an excellent source of energy, helps transport fat-soluble vitamins, insulates and protects interior tissues, and supports vital metabolic and cell growth processes. Additionally, adding fat to most of our diets is beneficial because lipids are necessary for a number of bodily activities (David *et al.*, 2014). There is no significant difference ( $P < 0.05$ ) between the samples A, B, & E and between the samples B & E and C & D. The average crude fat content of the samples is 6.34% which is similar to that of previous studies (Okaraonye *et al.*, 2009; Rambau *et al.*, 2016; Kamaruddin *et al.*, 2020).

Crude fibre is a dietary component essential to ruminant digestion, which has an impact on ruminant body weight and livestock output (Dhingra *et al.*, 2012; Wangchuk *et al.*, 2018). The majority of dietary fibre is made up of complex carbs, which are a little difficult to digest. Through bacterial enzymes, soluble polysaccharides may go through some metabolism in the small intestine and particularly in the large intestine, turning them into substances that help keep the colonic micro flora healthy and advantageous to digestion (Holscher *et al.*, 2017). Napier grass is ideal for ruminants because it reduces the amount of nutrients that can be digested in the rumen and increases the amount of nutrients that can be absorbed in the intestine after digestion. This increases animal productivity and raises the caliber of the meat and milk

**Table 2.** Agrobotanical characteristics of sewage wastewater irrigated Napier grass

	Sample-A	Sample-B	Sample-C	Sample-D	Sample -E
Plant Height (M)	2.10±0.13 <sup>c</sup>	1.92±0.49 <sup>d</sup>	1.47±0.41 <sup>e</sup>	2.195±0.56 <sup>a</sup>	2.05±0.32 <sup>b</sup>
Leaf Length (M)	1.36±0.068 <sup>a</sup>	1.16±0.081 <sup>b</sup>	1.13±0.063 <sup>c</sup>	1.026±0.17 <sup>d</sup>	1.1175±0.14 <sup>c</sup>
No of Leaves per plant	10.75±0.25 <sup>b</sup>	9.5±0.5 <sup>d</sup>	9.25±0.25 <sup>e</sup>	11.75±0.25 <sup>a</sup>	9.25±0.25 <sup>c</sup>
Fresh Weight of the harvested plant (kg)	0.294±0.016 <sup>bc</sup>	0.285±0.010 <sup>c</sup>	0.272±0.008 <sup>c</sup>	0.321±0.015 <sup>a</sup>	0.309±0.005 <sup>ab</sup>
Total dry matter of the harvested plant (kg)	0.073 ±0.003 <sup>c</sup>	0.069 ±0.001 <sup>c</sup>	0.062 ±0.001 <sup>d</sup>	0.108 ±0.003 <sup>a</sup>	0.098 ±0.003 <sup>b</sup>
Dry Matter %	24.83	24.21	22.80	33.64	31.72

Values of triplicate determinations (mean±SD; n=3) in the same row with different letters are significantly different ( $P < 0.05$ ).

produced. These bioactive substances may also serve as potential nutraceuticals for animal nutrition and animal products (Michalak *et al.*, 2021). The crude fibre of the studied ranges from 30.63 to 37.11%. There is no significant difference ( $P < 0.05$ ) between the samples B, C, D, & E and between the samples E, B & A. The average crude fibre of the sample is 34.41 % which is similar to that of previous studies (35.9 % Das *et al.*, 2016; 30.22 % Lounglawan *et al.*, 2014; 30.7 % Butterworth *et al.*, 1965; 31.98 % Kamaruddin *et al.*, 2020 and 32.4 % Senthil Murugan *et al.*, 2016). The nitrogen-free extract of the samples ranges from 33.85 to 35.96 in the studied samples. There is no significant difference between the samples in the NFE content. The average NFE content of the sample is 35.09 % which is similar to that of 37.34 % (Senthil Murugan *et al.*, 2016). The proximate composition of napier grass samples which is irrigated with wastewater have no negative significant difference when compared to the control sample.

#### Proximate composition of milk from cows fed Napier grass irrigated with wastewater

Proximate composition of cow milk is listed in Table 4. The crude protein of the samples ranges from 5.28 % to 5.85 %. The highest content of protein was in Sample A and lowest content was in Sample C, similar results were observed for crude protein content of Napier grass also. The average protein of the samples is 5.55 % which is similar/higher when compared to other crops fed such as (soybean meal-3.96 % and Spirulina -4.02 % Manzochhi *et al.*, 2020), faba bean-3.55 % and soy pass grain-3.65% Kand *et al.*, 2020), (Napier grass - 2.74% Harmini *et al.*, 2020), (Napier grass with silages -3.60% Mediksa *et al.*, 2016) . The crude fat of the samples ranges from 5.88 % to 6.23%. The highest content of crude fat was in Sample A and lowest content was in

Sample C. The crude fat of napier grass samples also followed similar pattern in which highest fat content was in sample A and lowest in sample C. The average fat of the samples is 5.97 % which is similar higher when compared to other crops fed such as (soybean meal-4.95 % and Spirulina -4.72 % Manzochhi *et al.*, 2020), faba bean-4.55 % and soy pass grain-3.83% Kand *et al.*, 2020), (Napier grass - 5.0% Harmini *et al.*, 2020), (Napier grass with silages -4.62% Mediksa *et al.*, 2016) . The highest protein and fat in the samples than the previous studies is due to the feed Napier grass which is irrigated with sewage water contains high protein, fibre, energy values, vitamins and minerals content in it. The solid not fat of the ranges from 10.07 % to 15.82 %. The average solid not fat content of the samples is 13.07 %. The highest content of solid not fat was in Sample C and lowest content was in Sample A. The total solid of the ranges from 16.3 % to 21.6 %. The average total solid content of the samples is 18.92 %. The highest content of total solids was in Sample C and lowest content was in Sample A. The both total solids and solid not fat contents in samples are higher than previous studies (11.94 % & 7.72 Bitew *et al.*, 2020) and (13.87 % & 8.68 % Mediksa *et al.*, 2016), this may be due to the highest protein and fat content of the samples. The electrical conductivity of the milk samples ranges from 2.26 to 2.42 mS/cm. The highest electrical conductivity was in the sample B and the lowest was in the sample D. The average electrical conductivity of the samples was 2.33 mS/cm, which is similar low to the previous studies (3.64 mS/cm Olaniyan *et al.*, 2023). The average electrical conductivity of the samples is lower than the previous studies due to the highest fat content in the samples than the previous studies. The moisture content of the samples ranges from 78.3 to 83.7 %. The average moisture content of the sample is 81.08 which is similar lower to that of

**Table 3.** Proximate composition of the sewage wastewater irrigated Napier grass

Parameters	Sample-A	Sample-B	Sample-C	Sample-D	Sample-E
Moisture (%)	6.93±0.37 <sup>b</sup>	8.45±0.72 <sup>a</sup>	8.86±0.85 <sup>a</sup>	8.67±0.65 <sup>a</sup>	7.78±0.45 <sup>ab</sup>
Crude Fibre (g/100g)	30.63 ± 2.23 <sup>b</sup>	35.73±2.89 <sup>ab</sup>	37.11±3.58 <sup>a</sup>	36.14± 3.31 <sup>a</sup>	32.46 ±2.35 <sup>ab</sup>
Crude Lipid (g/100g)	7.06 ± 0.45 <sup>a</sup>	6.40± 0.39 <sup>ab</sup>	5.51± 0.27 <sup>c</sup>	5.88 ±0.38 <sup>bc</sup>	6.85 ± 0.48 <sup>a</sup>
Ash Content (g/100g)	13.75 ± 1.23 <sup>a</sup>	12.51 ±1.05 <sup>ab</sup>	11.71±0.84 <sup>c</sup>	11.51±0.72 <sup>c</sup>	13.1± 0.96 <sup>ab</sup>
Total Nitrogen Content(g/100g)	2.02±0.4 <sup>a</sup>	1.85±0.2 <sup>c</sup>	1.69±0.18 <sup>d</sup>	1.83±0.3 <sup>c</sup>	1.93±0.27 <sup>b</sup>
Crude Protein(g/100g)	12.60 ± 2.56 <sup>a</sup>	11.51 ± 1.27 <sup>a</sup>	10.54±1.15 <sup>a</sup>	11.49 ±1.88 <sup>a</sup>	12.04 ± 2.16 <sup>a</sup>
NFE (g/100g)	35.96±6.47 <sup>a</sup>	33.85±5.6 <sup>a</sup>	35.13±5.84 <sup>a</sup>	34.98±6.29 <sup>a</sup>	35.55± 5.95 <sup>a</sup>
Energy (KJ/100 g)	1077.11±9.48 <sup>a</sup>	998.79±7.26 <sup>c</sup>	989.96±7.26 <sup>c</sup>	997.73±8.55 <sup>c</sup>	1043.48±8.59 <sup>b</sup>

Values of triplicate determinations (mean ± SD; n=3) in same row with different letters are significantly different ( $P < 0.05$ ).



previous studies 82.08 % (Dandare *et al.*, 2014) and 85.56 % (Madu *et al.*, 2020). There is no significant difference between the parameters such as pH, electrical conductivity, crude fibre, crude lipid, crude protein and energy values in the samples. There is a significant difference ( $P < 0.05$ ) between the samples in the total non-specific carbohydrates content. There is no significant difference ( $P < 0.05$ ) between the samples B & D in the total solids, solid not fat and moisture content. There is no significant difference ( $P < 0.05$ ) between the samples A, B, D, & E and E & B in the ash content of the samples. Both milk as well as napier grass samples had similar trend on crude protein and crude fat content, which implicates that the plant nutrients are reflecting in the milk quality. Proximate composition of cow milk fed with sewage wastewater irrigated napier grass is not affected when compared to irrigated with well water (control).

#### Macro-element composition of milk fed with sewage wastewater irrigated Napier grass

Macro-elements in particular, which are mineral components, found in milk, are crucial in creating the physical-chemical equilibrium of the colloidal system, which governs the milk's qualities. Macro-element is particularly important since it significantly affects the size of the casein particle. The type of animal feed and water that dairy cattle eat determines the composition of raw milk.

Calcium composition of milk samples is listed in the Table 5. The calcium content of the samples ranges from 1110 to 1176 mg/l. There is no significant difference ( $P < 0.05$ ) between samples A & B and B & D. The average calcium content of the samples is 1143.4 mg/l which is similar to the

previous studies of 1217 mg/L (Roger *et al.*, 2013), 1202.4 mg/l (Kapadiya *et al.*, 2016) and higher to the previous studies of 838 mg/l (Nogalska *et al.*, 2020) and 767 mg/l (Nogalska *et al.*, 2018). Calcium is a mineral that is most frequently linked to strong bones and teeth, but it also plays a critical role in blood clotting, assisting with muscular contraction, and regulating regular heartbeats and nerve activity (De Valle *et al.*, 2011). Magnesium composition of milk samples is listed in the Table 5. The magnesium content of the samples ranges from 114 to 128 mg/kg. There is no significant difference ( $P < 0.05$ ) between samples A & C, C & D, D & B, and B & E. The average magnesium content of the samples is 121.4 mg/l which is higher than the previous studies reported by different authors (103 mg/l- Roger *et al.*, 2013), (93 mg/l- Nogalska *et al.*, 2020 ; 95 mg/l- Nogalska *et al.*, 2018) and similar to that of the value (126.5 mg/l) which reported by Kapadiya *et al.* (2016). The body requires magnesium as a vitamin to remain healthy. Magnesium is essential for numerous bodily functions, including the production of protein, bone, and DNA as well as the control of blood pressure, blood sugar levels, and muscle and neuron function (Fiorentini *et al.*, 2021).

Sodium composition of milk samples is listed in the Table 5. The sodium content of the milk samples ranges from 487 to 564 mg/l. There is a significant difference ( $P < 0.05$ ) between the samples from different study areas. Further, the average sodium content of the samples is found to be 518.2 mg/l which is higher to the previous studies 310-356 mg/l (Roger *et al.*, 2013; Nogalska *et al.*, 2020; Nogalska *et al.*, 2018). Sodium is a flavouring agent, stabiliser, and binder for food. Due to the fact that bacteria cannot survive in high-salt environments, salt also

**Table 4.** Proximate composition of cow milk fed with sewage wastewater irrigated Napier grass

Parameters	Sample-A	Sample-B	Sample-C	Sample-D	Sample -E
pH	6.29	6.36	6.33	6.40	6.38
Electrical Conductivity mS/cm	2.33	2.42	2.29	2.26	2.38
Total Solids (TS) (%)	16.3±0.83 <sup>d</sup>	18.8±0.12 <sup>c</sup>	21.6±0.67 <sup>a</sup>	18.2±1.02 <sup>c</sup>	19.7±0.92 <sup>b</sup>
Solid Not Fat (SNF) %	10.07±2.56 <sup>d</sup>	12.92±1.46 <sup>c</sup>	15.82±3.34 <sup>a</sup>	12.17±2.28 <sup>c</sup>	13.78±2.5 <sup>b</sup>
Moisture (%)	83.7 ±0.83 <sup>a</sup>	81.2 ±0.12 <sup>b</sup>	78.4±0.67 <sup>d</sup>	81.8±1.02 <sup>b</sup>	80.3±0.92 <sup>c</sup>
Crude Fibre (g/100g FM)	0.19 ±0.03 <sup>a</sup>	0.34±0.12 <sup>a</sup>	0.31±0.18 <sup>a</sup>	0.28±0.15 <sup>a</sup>	0.24 ±0.08 <sup>a</sup>
Crude Lipid (g/100g FM)	6.23±1.76 <sup>a</sup>	5.88 ±1.34 <sup>a</sup>	5.78 ±2.67 <sup>a</sup>	6.02 ±1.26 <sup>a</sup>	5.92 ±1.58 <sup>a</sup>
Ash Content (g/100g FM)	0.83 ±0.11 <sup>a</sup>	0.74±0.08 <sup>ab</sup>	0.63 ±0.04 <sup>b</sup>	0.81±0.12 <sup>a</sup>	0.76±0.01 <sup>ab</sup>
Crude Protein(g/100g FM)	5.85±1.56 <sup>a</sup>	5.35±0.99 <sup>a</sup>	5.28±1.87 <sup>a</sup>	5.73±1.72 <sup>a</sup>	5.56 ±0.93 <sup>a</sup>
Total Non- Specific Carbohydrate	3.2 ±3.29 <sup>e</sup>	6.49 ± 2.65 <sup>c</sup>	9.6±5.43 <sup>a</sup>	5.36 ± 4.27 <sup>d</sup>	7.22± 3.52 <sup>b</sup>
Energy (KJ/100 g)	1783.79±6.78 <sup>a</sup>	1775.44±4.86 <sup>a</sup>	1775.6±9.30 <sup>a</sup>	1778.21±6.23 <sup>a</sup>	1777.62±5.11 <sup>a</sup>

Values of triplicate determinations (mean ± SD; n=3) in same row with different letters are significantly different ( $P < 0.05$ ).

serves as a food preservative. A little amount of sodium is needed by the human body to convey nerve impulses, contract and relax muscles, and keep the right ratio of water and minerals (Henney *et al.*, 2010). Potassium composition of milk samples is listed in the Table 5. The potassium content of the samples ranges from 1357 to 1578 mg/l. There is a significant difference ( $P < 0.05$ ) between the samples from different study areas. The average potassium content of the samples is 1461.6 mg/l which is on par with the previous studies 1384-1536 mg/l (Roger *et al.*, 2013; Nogalska *et al.*, 2018; Nogalska *et al.*, 2020). Potassium supports normal blood pressure maintenance, proper muscular function, and normal nervous system function (Stone *et al.*, 2016). Phosphorus composition of milk samples is listed in the Table 5. The phosphorus content of the samples ranges from 838 to 956 mg/l. For phosphorus, there is a significant difference ( $P < 0.05$ ) between the milk samples from different study areas. The average phosphorus content of the samples is 896.2 mg/l which is higher than the value (776 mg/l) reported by Roger *et al.* (2013). However, the phosphorus value, 880.8 mg/l is found to be similar to that of above mentioned experimental cow milk sample of Kapadiya *et al.* (2016). All tissues and cells require phosphorus for their growth, upkeep, and repair, as well as for the synthesis of DNA and RNA, the genetic building blocks. Additionally, other vitamins and minerals like vitamin D, iodine, magnesium, and zinc need phosphorus to be balanced and used (Kumar *et al.*, 2021).

### Heavy metals and microelements composition-water, soil, plant and cow milk samples

#### Aluminium

Aluminium content of all samples was listed in the Table 6. Aluminium is present only in the Ukkadam water sample but it is present in all the waste water irrigated soil and Napier grass and cow milk samples. The aluminium content present in

Ukkadam sample is 0.0281 mg per litre. The aluminium in the soil samples was from 0.00105 to 0.00991 mg/kg, 0.0419 to 0.248 mg/kg in the plant samples and from 0.0858 to 0.435 mg/l in the milk samples. The WHO maximum permissible limit of aluminium in water is 5 mg/l, the soil is 10 mg/kg and plant (animal feed) is 5 mg/kg. However, the aluminium content present in water, soil and plant is below the permissible limit. Cow milk naturally contains substantial amount of aluminium but our samples are found to contain less values when compared to the previous study is 2.93 micrograms/litre (Amer *et al.*, 2021). Although it is the most prevalent metal in the crust of the Earth, aluminium (Al) is not necessary for plant growth. The free metal cation of aluminium, Alaq (3+), is very physiologically reactive, and the biologically accessible aluminium is essentially poisonous and non-essential (Zhao *et al.*, 2018).

#### Arsenic

The total arsenic content in the water samples ranges from 0.022 to 0.0241 mg/litre, from 0.85 to 2.92 mg/kg in the soil samples, from 0.0248 to 0.134 mg/kg in the plant samples, and from 0.00492 to 0.00594 mg/l in the milk samples. Arsenic content of all samples was listed in the Table 6. The maximum permissible limit of arsenic in irrigation water is (0.10 mg l<sup>-1</sup>) by FAO. The maximum permissible limit of As content for agricultural soil recommended by the European Union is 20 mg/kg. The maximum permissible limit of As content in the plant (animal feed) and milk is 1.1 mg/l. The presence of arsenic content in water, soil, plant, and milk in the study area is found to be below the permissible limit. Every environment contains arsenic (As), which is extremely hazardous to all types of life (Alexander *et al.*, 2016).

#### Cadmium

The cadmium content in the water samples ranges from 0.0023 to 0.0395 mg/litre, from 0.43 to 3.12

**Table 5.** Macro-element composition of cow milk fed with sewage wastewater irrigated Napier grass (mg/L).

	Sample-A	Sample-B	Sample-C	Sample-D	Sample-E
Magnesium	128 ± 1.34 <sup>a</sup>	118 ± 1.76 <sup>cd</sup>	125 ± 3.36 <sup>ab</sup>	122 ± 2.58 <sup>bc</sup>	114 ± 3.82 <sup>d</sup>
Calcium	1152 ± 10.43 <sup>b</sup>	1145 ± 9.34 <sup>bc</sup>	1110 ± 7.32 <sup>d</sup>	1134 ± 8.78 <sup>c</sup>	1176 ± 10.23 <sup>a</sup>
Sodium	564 ± 3.2 <sup>a</sup>	498 ± 4.6 <sup>c</sup>	487 ± 2.87 <sup>d</sup>	504 ± 5.3 <sup>c</sup>	538 ± 4.42 <sup>b</sup>
Potassium	1552 ± 11.2 <sup>b</sup>	1357 ± 4.7 <sup>e</sup>	1437 ± 10.3 <sup>c</sup>	1578 ± 12.3 <sup>a</sup>	1384 ± 3.3 <sup>d</sup>
Phosphorus	907 ± 6.4 <sup>b</sup>	894 ± 8.3 <sup>c</sup>	838 ± 4.7 <sup>e</sup>	886 ± 8.8 <sup>d</sup>	956 ± 7.3 <sup>a</sup>

Values of triplicate determinations (mean ± SD; n=3) in same row with different letters are significantly different ( $P < 0.05$ ).

mg/kg in the soil samples, from 0.0017 to 0.01 mg/kg in the plant samples, and from 0.00085 to 0.00278 mg/l in the milk samples. The maximum permissible limit for Cd in water samples was 0.01 mg/l (Indian standards). The maximum permissible limit for Cd in soil samples was 3 to 6 mg/kg (Indian standards). The maximum permissible limit for Cd in food samples was 1.5 mg/kg (Indian standards). The presence of cadmium content in water, soil and plant samples in the study area was below the permissible limits. Interestingly, the content of cadmium in the above said milk samples are found to be much lower than the standard value (2.6 mg/l) of the International Milk Federation (IMF). Cadmium content of all samples was listed in the Table 6. Manures and pesticides are the main environmental sources of cadmium. The main sources of cadmium in surface waterways are household and industrial wastes. Cadmium is particularly hazardous when ingested through food and strongly binds to organic substances in soils (Kubier *et al.*, 2019).

### Chromium

The chromium content in the water samples ranges from 0.0013 to 0.0725 mg/l, from 34.3 to 68.5 mg/kg in the soil samples, from 0.0024 to 0.132 mg/kg in the plant samples and from 0.0012 to 0.00655 mg/l in the milk samples. The maximum permissible limit for Cr in water is 0.1 mg/l. the permissible limit of chromium in the soil is 100 mg/kg, in the plants is 1.30 mg/kg and in the milk is 0.05 mg/l (FAO/WHO). Chromium content of all samples was listed in the Table 6. The collected water soil, plant and milk sample concentration of chromium was recorded below the permissible limit set by WHO/FAO/Indian standards. Chromium is one of the most prevalent heavy metal pollutants in soil, sediments, and groundwater. The most prevalent and stable states in terrestrial environments are the trivalent ( $\text{Cr}^{3+}$ ) and hexavalent ( $\text{Cr}^{6+}$ ) states. It can exist in a variety of oxidation levels, from  $\text{Cr}^2$  to  $\text{Cr}^{6+}$  (Sharma *et al.*, 2022).

### Copper

The copper content in the water samples ranges from 0.0013 to 0.00085 mg/l, from 0.086 to 0.843 mg/kg in the soil samples, from 0.42 to 0.75 mg/kg in the plant samples, and from 0.0035 to 0.005 mg/l in the milk samples. The maximum permissible limit for Cu in water is 0.1 mg/l, in the soil is 36 mg/kg, in plant based green feeds is 10 mg/kg and in milk

is 0.5 mg/l by WHO/FAO/Indian standards. Copper content of all samples was listed in the table 7. In all the collected water, soil, plant (as feed source), and milk samples concentration of copper was recorded below the permissible limit as a heavy metal but the concentration in the milk samples is found to be adequate levels to meet out the human nutrient requirements. In addition to being useful in the enzymatic activities of biological systems, copper is a necessary metal for regular biological processes (Wang *et al.*, 2021).

### Iron

The iron content in the irrigation water samples ranges from 0.0378 to 0.242 mg/L, 0.00146 to 0.831 mg/kg in the soil samples, 0.286 to 0.86 mg/kg in the plant samples, and from 0.358 to 0.578 mg/l in the milk samples. The maximum permissible limit of iron in irrigation water is 0.5 mg/l (Indian standards). Though the concentration of iron in the water, soil, plant, and milk sample were recorded below the permissible limit set by WHO/FAO/Indian standards, the level of iron recorded in the milk samples is found to be on par with the human dietary recommendation suggested by WHO/FAO. Iron content of all samples was listed in the Table 7. In addition to being a crucial trace element for the oxidation of carbohydrates, proteins, and lipids as well as the production of hemoglobin, iron is also crucial for oxygen and electron transport in the body (Abbaspour *et al.*, 2014).

### Manganese

The Manganese content in the water samples ranges from 0.043 to 0.187 mg/l, from 116.7 to 296.5 mg/kg in the soil samples, from 18.9 to 38.4 mg/kg in the plant samples and from 0.19 to 0.38 mg/l in the milk samples. Mn is a necessary cofactor for numerous classes of enzymes, including oxidoreductases, transferases, ligases, and hydrolases, Mn is required for all living systems (Li *et al.*, 2018). The maximum permissible limit of manganese in water is 0.2 mg/l and in milk is 0.4 mg/l by WHO standards. The concentrations of Mn in all the samples are within the permissible limits. Manganese content of all samples was listed in the Table 7.

### Nickel

In the present study, the Nickel content in the water, soil, plant and milk samples which ranges from 0.0008 to 0.00381 mg/l, 34.23 to 71.7 mg/kg, 3.03 to 9.89 mg/kg, 0.0083 to 0.02 mg/l respectively.

Interestingly, there is no presence of nickel content in the control milk sample (E). Nickel content of all samples was listed in the Table 6. Nickel has been regarded as a trace element that is crucial for plant, human and animal health (Begum *et al.*, 2022). Nickel is considered to be an essential plant micronutrient because it acts as an activator of the enzyme urease. However, high concentrations of Ni in growth media severely retards seed germination of many crops. As concern the human nutrition, the nickel is playing a crucial role in the iron absorption, an essential micronutrient for hormone function and lipid metabolism (Shahzad *et al.*, 2018). The maximum permissible limit of Ni in irrigation water suggested by FAO is 0.20 mg/l and the maximum permissible limit of Ni in the soil is 75 mg/kg. Further, in general, the permissible limit of Nickel content suggested for plants by WHO is 10mg/kg. On the other hand, the maximum permissible limit of nickel in cow milk is 0.002 mg/l. As heavy metal point of view, the level of Ni in all the water, soil, plant and milk samples are found to be within the permissible limits suggested by WHO/FAO and Indian Standards.

#### Lead

In the present investigation, the observed concentration of lead in the irrigation water samples ranges from 0.0154 to 0.0456 mg/l, from 7.83 to

43.56 mg/kg in the soil samples, from 0.833 to 1.763 mg/kg in the plant samples and from 0.0013 to 0.0093 mg/L in the milk samples. Lead content of all samples was listed in the table 6. Although lead is not an essential element for plant growth but its get easily absorbed / adsorbed and gets accumulated in different plant parts. Lead accumulation in the soil inhibits germination of seeds and retards growth of seedlings. Its exposure at higher rates disturbs the plant water and nutritional relations such as rapid inhibition of root growth, stunted growth of the plant, chlorosis, and inhibition of enzyme activities, disturbed mineral nutrition and water imbalance and causes oxidative damages to plants. The maximum permissible limit of lead in irrigation water suggested by WHO is 0.5 -2.0 mg /L and suggested pattern of Indian standard is 0.1 mg/l. According to the Indian standards, the maximum permissible limit of lead in soil is 250-500 mg/kg and in plants is 0.1-10 mg/kg respectively. Further, as per the Indian standards, the maximum permissible limit of lead in cow milk is 0.0002 mg/l. In all the respective samples, the lead content is found to be within the permissible limits.

#### Titanium

The content of titanium in the soil samples ranges from 0.0109 to 0.995 mg/kg and in plants from 0.00012 to 0.00194 mg/kg. Titanium content of all

**Table 6.** Heavy metals and microelements composition-water, soil, plant and cow milk samples

Samples	Al	As	Ba	Cd	Cr	Ni	Pb	Sn
A-water (mg/l)	-	0.0241	0.0621	0.0395	0.0725	0.00123	0.0188	0.00623
A-soil (mg/kg)	0.00991	2.36	-	3.12	63.7	43.5	17.67	0.00535
A-plant (mg/kg)	-	0.0903	0.00602	0.01	0.0056	4.79	0.976	-
A-milk (mg/l)	0.0858	0.00594	-	0.00265	0.00205	0.0083	0.0056	-
B-water (mg/l)	0.0281	0.033	-	0.0341	0.0528	0.00381	0.0456	0.00389
B-soil (mg/kg)	0.0013	2.92	-	2.85	55.3	71.7	43.56	0.00421
B-plant (mg/kg)	-	0.134	0.00598	0.00874	0.0399	9.89	1.763	-
B-milk (mg/l)	0.135	0.00576	-	0.00198	0.00264	0.02	0.0093	-
C-water (mg/l)	-	0.022	0.036	0.0141	0.0689	0.00198	0.0266	0.00557
C-soil (mg/kg)	0.00421	2.83	-	1.97	59.4	54.89	25.12	0.00392
C-plant (mg/kg)	-	0.0419	0.00605	0.00857	0.0591	5.78	1.345	-
C-milk (mg/l)	0.435	0.00592	-	0.00278	0.00407	0.0088	0.0048	-
D-water (mg/l)	-	0.015	-	0.0221	0.0852	0.00358	0.0154	0.00412
D-soil (mg/kg)	0.00385	1.87	-	2.08	68.5	66.23	13.84	0.00407
D-plant (mg/kg)	-	0.122	0.00601	0.00763	0.132	6.56	0.865	-
D-milk (mg/l)	0.171	0.00565	-	0.002	0.00655	0.0092	0.0026	-
E-water (mg/l)	-	0.002	-	0.0023	0.013	0.0154	-	0.00285
E-soil (mg/kg)	0.00105	0.85	-	0.43	34.3	34.23	7.83	0.0039
E-plant (mg/kg)	-	0.0248	0.00596	0.0017	0.0024	3.03	0.833	-
E-milk (mg/l)	0.409	0.00492	-	0.00085	0.0012	-	0.0013	-

Al -Aluminium, As -Arsenic, Ba -Barium, Cd -Cadmium, Cr -Chromium, Ni -Nickel, Pb -Lead and Sn -Tin.

samples was listed in the Table 7. Application of Titanium present in the soil stimulate plant growth, chlorophyll content, enzyme activities and uptake of major and minor nutrients in a species-specific manner. However, Ti is generally present in most of the plants in relatively low concentrations but there is no evidence about the essential participation of naturally occurring Ti in plant growth and metabolism (Lyu *et al.*, 2017). Nonetheless, the level of Titanium is well below the detectable limit in respective water and milk samples.

### Zinc

In the experimented samples such as irrigation water, soil, plant and milk, the concentration of zinc is found to be the ranges from 0.0343 to 0.456 mg/l, 72.44 to 250.2 mg/kg, 29.55 to 48.6 mg/kg and 0.00248 to 0.00667 mg/L respectively. Zinc content of all samples was listed in the Table 7. Zinc is one of the crucial trace elements and plays a critical role in growth, development, and defense and it activates enzymes that are responsible for the synthesis of certain proteins in plants. However, a low level of zinc can stunt growth, minimize reproductive sites, and can reduce yields in all crops. Nevertheless, an organism may become poisonous from higher zinc doses. Zinc is a relatively low concentration in surface water due to its limited mobility from the site of rock weathering or from natural sources, and

it plays a vital role in protein synthesis (Prashanth *et al.*, 2015). As per the WHO-FAO standards and Indian standard, the maximum permissible limit of zinc in irrigation water is 5 mg /l and 2 mg/l respectively. According the Indian standards, the maximum permissible limit of zinc in the soil, plant and milk samples are 300-600 mg/kg, 50 mg/kg and 0.006 mg/l respectively. Though the zinc is found to be a heavy/toxic metal, its concentration present in all the investigated samples are within the permissible limits.

Among the different experimented samples, Vanadium is present only in soil samples and there is no permissible limit for tin, vanadium and strontium presence in the soil samples. Strontium present only in soil and plant samples. Barium is present only in the water samples of Nanjundapuram and Vellalore which are below the permissible limit (WHO). The heavy metals such as antimony, selenium, thallium, cobalt, silver, molybdenum, and beryllium are found to be below the detectable limit in all the experimented samples such as irrigation water, soil, plant and milk samples. The heavy metal and micro-macro element contents in sewage wastewater irrigated samples is present higher than the control but its within the permissible limits when compared with the WHO, FAO and Indian standards.

Because the milk samples' heavy metal content is

**Table 7.** Heavy metals and microelements composition-water, soil, plant and cow milk samples

Samples	Sr	Ti	V	Cu	Fe	Mg	Zn
A-water (mg/l)	-	-	-	0.00047	0.242	0.187	0.0709
A-soil (mg/kg)	0.274	0.995	0.076	0.321	0.00286	245.8	109.7
A-plant (mg/kg)	-	0.00553	0.00194	0.59	0.286	34.6	33.76
A-milk (mg/l)	-	-	-	0.0035	0.554	0.31	0.00308
B-water (mg/l)	-	-	-	0.0004	0.147	0.157	0.128
B-soil (mg/kg)	0.46	0.133	0.0386	0.447	0.00284	196.4	176.8
B-plant (mg/kg)	-	0.00748	0.00141	0.68	0.453	30.24	42.4
B-milk (mg/l)	-	-	-	0.0043	0.358	0.29	0.00588
C-water (mg/l)	-	-	-	0.00023	0.147	0.133	0.0831
C-soil (mg/kg)	0.806	0.672	0.0665	0.439	0.00236	180.6	123.5
C-plant (mg/kg)	-	0.00627	0.00111	0.64	0.286	24.3	37.2
C-milk (mg/l)	-	-	-	0.0041	0.495	0.23	0.00501
D-water (mg/l)	-	-	-	0.00085	0.11	0.201	0.456
D-soil (mg/kg)	0.243	0.677	0.052	0.843	0.00831	296.5	250.2
D-plant (mg/kg)	-	0.06085	0.00139	0.75	0.86	38.4	48.6
D-milk (mg/l)	-	-	-	0.005	0.578	0.38	0.00667
E-water (mg/l)	-	-	-	0.00013	0.0378	0.043	0.0343
E-soil (mg/kg)	0.213	0.0109	-	0.086	0.00146	116.7	72.44
E-plant (mg/kg)	-	0.00409	0.00012	0.42	0.768	18.9	29.55
E-milk (mg/l)	-	-	-	0.0037	0.439	0.19	0.00248

Sr -Strontium, Ti -Titanium, V -Vanadium, Cu -Copper, Fe -Iron, Mg -Manganese and Zn -Zinc.

within acceptable bounds and they also meet other crucial milk quality criteria as proximal, macro- and micromineral content and heavy metal composition, it is safe to consume the milk. There is no bioaccumulation of heavy metals in the soil to the plants and in the plant to the cow milk. This is mostly because of a process called phytoextraction, which involves extracting heavy metals from polluted soil and water and absorbing them through plant roots (Osmana *et al.*, 2020). This might be because heavy metals are retained by plant roots due to insolubilization or cell compartmentalization, preventing their release to the xylem (Page *et al.*, 2015; Juel *et al.*, 2021). As a result, the roots will retain heavy metals considerably more so than the shoots. Napier grass has a harvesting time of 6 to 7 weeks, but it is not until the 8 weeks that the accumulation of heavy metals in shoots is significant. Plants release a huge range of metabolites from their roots into the rhizosphere to manage nutrient bioavailability and scope with environmental trace element stresses (Ma *et al.*, 2022). Therefore, in the present study, less concentration of heavy metals and various nutrient enrichment which registered in the irrigation water, soil, plant and cow milk samples from the urban, semi-urban and rural areas without any pollution and hazardous.

### CONCLUSION

From the above study, it is clear that domestic sewage wastewater is highly suitable for the cultivation of the Napier grass due to the proper mitigation measures taken by the government, but the periodical monitoring of the heavy metals in sewage water and soil has to be done to ensure the quality. Further, the cultivation of napier grass with domestic sewage water leads to reduction in the usage of fresh water, extraction of groundwater resources for irrigation and helps to achieve a zero agro-waste ecosystem without any metallic pollution. Particularly, the Napier grass as an effective and potential feed for livestock management leads to the production of cow milk which is economically supportive for the farmers and the society. Moreover, the present research investigation indicates that growing Napier grass with irrigation of domestic sewage water is a simple operation that doesn't require specialised irrigation costs, fertilizer application as this sewage water itself act as a potential liquid organic fertilizer and

also less labour involvement from planting to production. Further, the farmer should harvest the napier grass within 50 days from the previous cuttings because only there is a minimal absorption level of heavy metals and more production of digestible fibres. The farmers also reported that a magnificent two-fold increase in milk yield from 3 to 6 litres per day, after switching from dry feed to Napier grass fodder. Hence the cultivation of Napier grass with irrigation of properly mitigated domestic sewage wastewater and fed to the cow for milk production in urban/semi-urban agroecosystem is found to be one of the potential, cost-effective and environmentally secured way for sustainable feed production and consumption. Particularly, the small scale farmers will be more benefited without generating any pollutants and waste in the agro ecosystem. Recently the Government of the India has initiated as a public awareness program among the small farmers in semi-urban area those who involved in cow milk production has facing the problem of aflatoxin in feed and milk with using the long term stored wheat and rice straw feed source. In this context scientific community has suggested that the Napier grass / elephant grass (*Pennisetum purpureum*) cultivation in semi-urban area under wastewater irrigation and offering as alternate feed source for the milking cows is found to be natural based and cost efficient solution for the eradication of aflatoxin problem. On the other hand, it is found to be one of the cost-effective and nature based water recycling methods. Even though, the presence of heavy metals in the milk samples where within the permissible limits, the quality and long-term effects of consumption of this milk by humans needs to be continuously monitored for avoiding the health related risk.

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### Conflict of interest

No conflict of interest.

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