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Chemical and Nutritional Characterization of the Different Organs of Taif's Rose (*Rosa damascena* Mill. var. *trigintipetala*) and Possible Recycling of the Solid Distillation Wastes in Taif City, Saudi Arabia

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Abstract: The objective of the current study was to examine the chemical composition and biological functions of the various Taif's rose (TR) organs and floral solid distillation wastes (SDW). Additionally, it assessed the SDW's potential use in animal feed and potential health applications. For chemical and biological analyses, the plant stems, leaves, and flowers as well as the SDW of TR were gathered from four farms in the Al-Shafa highland region of Taif, Saudi Arabia. The highest levels of cardiac glycosides, flavonoids, and phenolics were found in the flowers (7.66 mg securiaside g^{-1}, 16.33 mg GAE g^{-1}, and 10.90 mg RUE g^{-1}, respectively), while the highest carbohydrate and alkaloid contents were found in the TR leaves (2.09% and 9.43 mg AE g^{-1}, respectively) with no significant differences from the SDW. Quercetin, apigenin, and rutin flavonoids, as well as isocorydine and boldine alkaloids, were found in larger concentrations in the flowers and floral SDW than in the leaves and stems. The various TR flower extracts were effective against Gram-negative and -positive bacteria but had no effect on fungal strains, but the SDW's methanol extract was only effective against fungi. The plant stem had the highest N, K, and Mg contents (138, 174, and 96.12 mg kg^{-1}, respectively), while the leaves had the highest P and Ca values (6.58 and 173.93 mg kg^{-1}, respectively). The leaves had the highest contents of total carbohydrates and acid detergent fibre (59.85 and 3.93%, respectively), while the stems had the highest total protein and acid detergent fibre (8.66 and 24.17%, respectively), and the SDW had the highest fats and crude fiber (0.57 and 36.52%, respectively). The highest amounts of digestible crude protein, gross energy, and total dissolved nutrients (TDN) (4.52% and 412.61 Mcal kg^{-1}) were found in the plant stem and flowers, respectively. The results of the current experiment showed that the TDN contents of the various organs and the SDW of TR are suitable for mature dry gestating beef cows. It was determined that, in addition to the SDW's potential usage as an ingredient in animal feed, various plant parts and TR's SDW can be utilized for a variety of medical reasons.

Keywords: macronutrients; phytochemistry; distillation wastes; rose flowers; biological activity

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1. Introduction

Because of the superior quality of its essential oils, Taif's rose (*Rosa damascena* Mill. var. *trigintipetala*), a Rosaceae plant, is one of the most widely produced commercial commodities [1]. The mature plant (four years or older) is a shrub that grows to a height of about 2.5 m and blooms once a year (in May or June), producing 500–600 flowers [2,3]. TR may be found growing in temperate and subtropical areas between 300 and 2500 m

above sea level [4]. It is grown for commerce among other places in Saudi Arabia, Egypt, Turkey, Morocco, Bulgaria, Iran, France, China, and India [5]. In several Taif Governorate locales, it is also one of the significant aromatic plants grown for use in the perfume, medicinal, and culinary sectors [6]. TR has strong antioxidant, antidiabetic, anti-HIV, antibacterial, anti-inflammatory, and cardiotonic properties since it contains a variety of phytochemical components, including alkaloids, phenolic acids, flavonoids, and other phenolic compounds [1,7–10]. It has historically been used to treat menstrual bleeding, inflammation, respiratory issues, depression, constipation, and chest pain [11,12].

TR is thought to be one of the most essential aromatic crops producing highly regarded essential oil [13,14]. The most popular TR products are rose flowers, rose water, essential oils, vegetative wastes, and distillation wastes [10]. Because the yield of rose oil is so low compared to the amount of rose flower utilized, a considerable quantity of rose flowers must be grown in order to produce enough rose water and oil [15]. In total, 4000 kg of rose petals are required for every kilogram of rose oil [16]. Therefore, in order to satisfy the needs, a huge amount of rose flowers are used in the distillation process, resulting in a large amount of rose wastes, which is a concern. Furthermore, according to Galal et al. [17], a lot of rose flowers must be used to make a tiny amount of rose water, and this process generates a lot of semisolid waste that could harm the environment and results in a significant amount of resource waste in addition to environmental pollution [18].

Natural sources of polyphenols, flavor compounds, polysaccharides, proteins, etc. can be found in the solid waste from TR oil distillation [19,20]. Large amounts of trash can be produced if there are insufficient essential oils in the raw materials (flowers, stems, leaves, etc.) [21]. Even though distilleries often discard the wastes, these routine practices may upset the ecological balance in the area and result in the loss of valuable physiologically-active compounds (polyphenols, fragrance molecules, and polysaccharides) that are present in the trash [22]. Distillation wastes can be composted and used as animal feed [3]. A Taif factory produces roughly 7000 L of liquid waste after utilizing 2500 kg of rose flowers to extract the oil. One oil factory may repeat this extraction procedure twice or three times daily for 45 days during the flowering season. Therefore, in addition to the solid distillation wastes (SDW) of the flower leftovers, roughly 63,000–94,500 L of liquid waste are created from 225,000–337,500 kg of rose flowers each season. This is a significant amount of oil distillation wastes, and if they are dumped into the environment from just one factory, they might lead to a number of environmental issues [23]. As a result, recycling these wastes is crucial for maintaining environmental safety. According to the previous studies on TR organs, the leaves and stems can be used for different medicinal purposes [1,10], animal forage and organic fertilizers [17], and phytoremediators [4].

The majority of studies on TR have concentrated on the growth methods, the timing of the harvest, and the physical and chemical qualities of the oil [23]. However, pharmacological investigations have demonstrated the diverse health benefits of TR's vegetative leaves and stems [1] and reproductive flowers [24], which are mostly attributable to their abundant polyphenolic components. Different portions of the TR plant yield a variety of phytochemicals, including flavonoids, glycosides, alkaloids, and other phenolic compounds [1,10,25]. Additionally, macronutrients contribute significantly to the high yield and excellent quality of TR oil production [26]. The flower output and essential oil quality of TR plants depend on their nutritional balance [27]. Galal et al. [17] looked into the chemical make-up of the main macronutrients in the TR pruning wastes. The goal of the current study was to better understand the chemical composition of the various TR organs as well as the solid byproducts of flower distillation and their biological activity. In addition, it assessed how the distillation wastes might be used to improve human health when compared to various plant parts and how their disposal could lessen environmental pollution. Thus, the study investigated the inorganic and organic nutrients, nutritive value, and the phytochemical constituents of the different organs of TR as well as the SDW. Such a study can help in the recycling of TR wastes and their multipurpose utilization including for medicinal use, animal fodder, and organic fertilizers.

2. Materials and Methods

2.1. Plant Sampling

The various TR parts (stem, leaves, and flowers) were gathered from four rose farms in Taif City, Saudi Arabia's Al-Shafa Highland. The residual plant materials and the distillation wastewaters are the two main byproducts of oil distillation that are produced as industrial wastes [28]. The rose petals from each farm were combined with water in a weight-to-volume ratio of 1:10 to extract the oil [29]. Following the extraction process, the solid distillation wastes (SDW) from each farm were collected as triplicates. In addition to the SDW, three composite samples of each plant organ were also collected from each farm (N = 48) during the harvesting season in May 2021 for additional chemical and biological investigations.

2.2. Sample Preparation and Quality Analysis

Before being homogenized in a planetary high-energy mill with a hardened chromium steel vial for plant analysis, three composite samples of TR (leaves, stems, flowers, and SDW) were air-dried at room temperature in the shade. A 250 g sample of plant powder was shaken in 1000 mL of ethanol for 24 h at room temperature using an orbital shaker, and the extract was then filtered through Whatman No 1 filter paper. Through the use of an evaporator, the filtrate was condensed to dryness at 40 °C under reduced pressure. For the examination of alkaloids, phenolic acids, flavonoids, and cardiac glycosides, the extract was kept between 2 and 8 °C. The chemical reagents that were employed included HCl, ethanol, Baljet's solution, picric acid, NaOH), $AlCl_3$, methanol, Folin reagent, $NaHCO_3$, formic acid, acetonitrile, glacial acetic acid, diethylamine, dimethyl sulfoxide, ketoconazole, and gentamicin.

2.3. Phytochemical Analysis

2.3.1. Determination of Soluble Carbohydrates

The total soluble carbohydrates were determined using the anthrone technique [30]. In total, 100 mg of the TR powdered sample were hydrolyzed with 5 mL of 2.5 N HCl in a boiling-water bath for 3 h. Before adding sodium carbonate to neutralize the acid-digested sample, it was cooled to room temperature first. The resultant volume was diluted to 100 mL with distilled water and centrifuged for 15 min at 5000 rpm. The supernatant was then collected in order to calculate the total amount of soluble carbohydrates.

2.3.2. Determination of Cardiac Glycosides

In order to measure cardiac glycosides, the method of Solich et al. [31] and Tofighi et al. [32] was employed. A 10% ethanol extract was combined with 10 mL of freshly-made Baljet's solution (95 mL of 1% picric acid with 5 mL of 10% NaOH) in order to identify cardiac glycosides. A spectrophotometer (CECIL CE 1021) was used to measure the absorbance at 495 nm after the solution had been left to stand for an hour.

2.3.3. Determination of Total Flavonoid Contents (TFC)

The TFC of the sampled plant organs as well as the SDW were calculated using the methods given by Tofighi et al. [32]. The plant sample was extracted using a 20 mL water–ethanol solution at a 60% (v/v) concentration for 60 min at reflux (80 °C; pH = 5.06). The extract was filtered when it had reached room temperature, and the residue was then extracted once more using the same procedures. The volume was increased to 50 mL of water–ethanol solution at 60% (v/v) after the hydroalcoholic extract and the re-extract were combined (stock solution). A portion of the stock solution was transferred to a 10-mL volumetric flask and mixed with methanol to make the stock solution volume (blank solution). A new 10-mL volumetric flask was used, and a second aliquot of the stock solution was placed into it. The flask was then filled with 2% $AlCl_3$ and methanol was used to bring it to volume (test solution). The test solution's absorbance at 430 nm was evaluated after 25 min in comparison to a control solution. The average of three measurements was

used to calculate the rutin content of the TFC herbal material. The following formula was used to calculate the flavonoid content (mg g^{-1}) of herbal material (corrected for moisture content): TFC herbal material is calculated as (TFC tested solution 1.25 × 50)/(w-ld), where TFC test solution is the total amount of flavonoids in the test solution (mg mL^{-1}), 1.25 is the dilution factor, 50 is the volume of the stock solution (mL), w is the weight of the herbal material (g), and l d is the herbal material's loss during drying.

2.3.4. Determination of the Total Phenolic Compounds

Using a spectrophotometric technique, the amount of phenolics in the plant's ethanol extract was assessed [32,33]. Amounts of 0.5 mL of ethanol extract, 2.5 mL of 10% Folin reagent Ciocalteu's dissolved in water, and 2.5 mL of 7.5% $NaHCO_3$ made up the reaction mixture. The blank was created using 0.5 mL of methanol, 2.5 mL of water-dissolved Ciocalteu's 10 percent Folin reagent, and 2.5 mL of $NaHCO_3$ at a 7.5 percent concentration. The samples were then incubated for 45 min at 45 °C in a thermostat. Using a spectrophotometer, the absorbance at 765 nm was measured (CECIL CE 1021). For each experiment, samples were prepared in triplicate, and the mean absorbance value was calculated. The same process was used to produce the calibration curve for the standard gallic acid solution. Per gram of dry weight, the amount of phenolics was calculated as milligrams of gallic acid equivalent (GAE) (DW).

2.3.5. Estimation of Phytochemical Compounds Using HPLC

For the separation, identification, and quantification of flavonoids, phenolic acids, and other phenolic chemicals in TR plant samples, HPLC-MS techniques are frequently utilized. The HPLC-MS system (Agilent 1100: Agilent Corp., Palo Alto, CA, USA) is made up of a single quadrupole MS detector with an ion source, a photodiode array detector, a quaternary pump, and UV/Vis detectors (ESI). Using a gradient solvent system with a flow rate of 1.0 mL min^{-1} and a 0.1% formic acid solution, flavonoids were separated in 70 min. They were then detected at 280 nm and identified by ESI-MS [34]. With a gradient mobile phase consisting of water/acetonitrile/glacial acetic acid (980/20/5, *v*/*v*/*v*, pH 2.68) and acetonitrile/glacial acetic acid (1000/5, *v*/*v*), phenolic acid was separated in 60 min [35]. Additionally, alkaloids were examined by HPLC using 0.2% diethylamine and 0.16% formic acid as solvent system A and 0.2% diethylamine and 0.16% formic acid in acetonitrile as solvent system B (0 min, 80:20 (A:B); 5 min, 80:20; 20 min, 60:40; 25 min, 0:100). The column used had a flow rate of 1.0 mL min^{-1} and was a 5 µm, 250 mm × 4.6 mm GraceSmart RP18 (Grace Vydac, Hesperia, CA, USA). At 226 nm, the peaks were discovered.

2.4. Biological Activity

2.4.1. Preparation of Extracts

Since the biological activity of TR leaves and stems had already been established [1], 250 g of plant powder from the flowers and SDW were steeped in 1.5 L of 95% ethanol and methanol before being boiled in both cold (about room temperature) and warm (50 °C) water for 5 days. To ensure a uniform infusion, the mixture was blended every day. After 5 days, Whatman filter paper No. 1 was used to filter the extract. The filtrate was dried in a rotary evaporator at a temperature of 60 °C. The dried extract was stored in sterile glass vials at −20 °C until use [36].

2.4.2. Microorganisms Used

One Gram-positive bacterial strain (*Bacillus subtilis*), two Gram-negative bacterial strains (*Escherichia coli* and *Proteus vulgaris*), and two fungal strains (*Aspergillus fumigatus* and *Candida albicans*) were obtained from Faculty of Science at Al-Azhar University in Cairo, Egypt. In nutrient agar and malt extract, respectively, the bacterial and fungal strains were cultivated.

2.4.3. In Vitro Evaluation of the Antimicrobial Activity

By modifying the agar disc well diffusion method [37], an antimicrobial susceptibility test was carried out. Antimicrobial activity was gauged by measuring the diameter of inhibitory zones. As antibacterial agents, plant extracts were tested on bacterial isolates. All bacterial and fungal isolate inoculum suspensions were dispersed across the surface media. The medium was drilled with holes that were 6 mm in diameter using a 6-mm cork borer. Dimethyl sulfoxide (DMSO) was used to process the dried plant extracts into a final extract with a 10 mg/mL concentration. In total, 100 L of plant extract was placed in the well of each plate. For bacterial growth, the inoculated agar plates were incubated for 24 h at 37 °C and for 48 h at 28 °C. Measurements of the inhibitory zones brought on by active extract components were made after 24–48 h of incubation. The investigations were carried out in duplicate, and an established scale was used to evaluate the inhibitory zone [38]. For fungus, gentamicin (MIC = 4 g mL^{-1}) was utilized as a control therapy, whereas ketoconazole antibiotic (MIC = 100 g mL^{-1}) was employed for bacteria.

2.5. Nutritional Analyses

2.5.1. Mineral Nutrients

Three composite samples of TR plants from each farm (N = 48) were pulverized separately in a metal-free plastic mill, passed through a 2-mm mesh size, and then stored in labeled plastic containers. The samples included leaves, stems, flowers, and SDW. For the measurement of inorganic nutrients, a sulphuric and perchloric acid mixture was used to digest 1 g of each milled plant sample [39]. At the Ecology Laboratory, Faculty of Science, Helwan University, Cairo, Egypt, the determination of N, P, K, Ca, Mg, and Na was performed for the extracts using an Agilent 4210 MP-AES (Microwave Plasma-Atomic Emission Spectrometer, Agilent Inc., Suwon, Korea). The operational processes and instrumental settings were modified in accordance with the user guide provided by the manufacturer. For each element, the final concentrations were given as mg kg^{-1} biomass dry matter.

2.5.2. Organic Nutrients

The ground plant samples were ignited for three hours at 550 °C in a muffle furnace to determine the amount of ash present. The Kjeldahl method [40] was used to assess total nitrogen, and its content was utilized to compute total protein (TP) by multiplying its percentage by 6.25. Allen [39] reported that the total lipids or fats represented by ether extract (EE) were determined by diethyl ether extraction of the plant materials using a Soxhlet extractor. Allen [39] claims that after chemically digesting and solubilizing the other existing components, the crude fibre (CF) is measured gravimetrically. According to Le Houérou [41], the nitrogen-free extract (NFE, or total carbohydrates) was determined in the plant samples as follows: NFE (% DM) = 100 − (TP + CF + fat + ash). According to Goering and Van Soest [42] and Van Soest et al. [43], the fiber portions of cell walls were determined as neutral detergent fiber (NDF), acid detergent fiber (ADF), and acid detergent lignin (ADL).

2.5.3. Nutritional Value

According to NRC [44], the digestible crude protein (DCP) was calculated as follows: DCP percentage (% DCP) = 0.85 CP − 2.5. According to NRC [45], the digestible energy (kcal kg^{-1} DM) was determined as follows: gross energy times 0.76 equals digestible energy (DE). The metabolizable energy (kcal kg^{-1} DM) was determined as follows [44]: digestible energy multiplied by 0.82 equals metabolic energy (ME). According to NRC [44], the following formula was used to determine the net energy (kcal kg^{-1} DM): metabolizable energy times 0.56 equals net energy (NE). The caloric values were expressed in Mcal kg^{-1} DM. The total digestible nutrients (TDN) were computed as follows NRC [44]: % digestible energy divided by 44.3 equals total digestible nutrients (%).

2.6. Statistical Analysis

Following a normality check of the data, one-way analysis of variance (ANOVA I) was used to examine the variations in the chemical composition of the plant's various organs [46]. A post hoc test (Duncan's test) was employed when there were significant differences between the different samples.

3. Results and Discussion

3.1. Phytochemical Analysis

It is crucial to identify phytochemical components in order to evaluate the nutritional and therapeutic value of plants [47]. The TR plants' phytochemical components showed substantial differences ($p < 0.001$) among the various plant organs (Table 1). According to the qualitative phytochemical examination of TR's various organs, carbohydrates, glycosides, alkaloids, flavonoids, and phenolic compounds were found. The same species' petals [48,49], and leaf and stem pruning wastes [1] also revealed similar results. Carbohydrates are substances with a lot of energy that show how much food and energy plants have stored and offer the cellular building blocks for those plants [50]. They are used to monitor a plant's capacity to rebound following pruning [51]. TR leaves were observed to have much higher carbohydrate and alkaloid concentrations than other plants (2.09 percent and 9.43 mg AE g^{-1}, respectively), but lower levels of cardiac glycosides (3.61 mg securiaside g^{-1}). The drought stress of rose plants before and after pruning may be the cause of the elevated carbohydrate content in the leaves [6]. Additionally, the reduced ability to use carbohydrates as an energy source, a component of new cells and tissues, or an osmolyte for the cells, may be the cause of the accumulation of carbohydrates [37].

Table 1. Phytochemical constituents (mean ± SD, n = 12) of the different organs as well as the solid distillation wastes of Taif's rose collected from Al-Shafa highlands in Taif City, Saudi Arabia. Maximum and minimum values are underlined.

Organ	Carbohydrates %	Cardiac Glycosides mg Securiaside g^{-1}	Phenolics mg GAE g^{-1}	Flavonoids mg RUE g^{-1}	Alkaloids mg AE g^{-1}
Leaves	2.09 ± 0.71 a	3.61 ± 0.59 c	9.81 ± 1.79 b	6.33 ± 1.52 b	9.43 ± 1.17 a
Stem	0.85 ± 0.08 b	4.84 ± 0.63 b	5.45 ± 1.55 c	7.46 ± 1.16 b	5.70 ± 1.21 b
Flower	0.66 ± 0.08 b	7.66 ± 0.20 a	16.33 ± 0.10 a	10.90 ± 0.20 a	3.44 ± 0.14 c
Flower wastes	0.56 ± 0.04 b	5.63 ± 0.16 b	14.36 ± 2.11 a	9.80 ± 0.26 a	2.36 ± 0.10 c
F-value	19.73 ***	38.54 ***	55.75 ***	13.98 ***	23.98 ***

Means in the same column followed by different letters are significantly different at $p < 0.05$, according to Duncan's HSD test; ***: $p < 0.001$.

Additionally, the TR flowers contained the most cardiac glycosides (7.66 mg securiaside g^{-1}). This figure was higher than what Galal et al. [1] found in the TR leaf and stem pruning wastes. Cardiac glycosides are a particular class of secondary metabolite that have historically been used to increase cardiac contractile force in people with congestive heart failure or cardiac arrhythmias [52]. Their content was higher than the 2.98–5.69 mg g^{-1} found in TR's leaf and stem pruning wastes [1] but lower than the 9.5–15.2 mg g^{-1} and 9.07–21.09 mg g^{-1} found in *C. procera* and *Aloe* spp. ([52,53], respectively). Moreover, the SDW had the lowest levels of soluble carbohydrates and alkaloid contents (0.56% and 2.36 mg AE g^{-1}, respectively), whereas the stem tissues had the lowest levels of phenolics (5.45 mg GAE g^{-1}).

The TR flowers also had the highest levels of flavonoid and phenolic compounds (16.33 mg GAE g^{-1} and 10.90 mg RUE g^{-1}, respectively), with no discernible changes from the SDW. In the same species, Galal et al. [1] and Liu et al. [18] recorded 12.41 and 386.4 mg g^{-1} of phenolic content in the leaf pruning wastes and flower residue, respectively. These results suggest that the TR flowers and SDW have better pharmacological (antioxidant, anti-ageing, whitening, and anticancer) actions than the TR leaves and stems. Additionally, numerous studies have shown how TR's high phenolic and flavonoid con-

tents contribute to its potent antibacterial and disinfection properties [24]. Moreover, the TR flower's flavonoid concentration was higher than the 9.33 mg g^{-1} found in the same species' stem and leaf pruning wastes [1].

3.2. HPLC of Phytochemical Compounds

The HPLC analysis of the flavonoids, and phenolic and alkaloid compounds showed that the flowers and flower SDW of TR plants had lower contents of the separated compounds (except quercetin, apigenin, and rutin flavonoids, and isocorydine, and boldine alkaloids) than the leaves and stems (Table A1). Quercetin, apigenin, luteolin, chrysoeriol, rutin, kaempferol, and avicularin were the seven flavonoid compounds that were isolated and identified (Table A1 and Figure 1). These chemicals have potential antioxidant, anti-inflammatory, and antimicrobial properties as reported by Dahat et al. [54], who discovered that quercetin and its glycoside rutin have been identified in extracts demonstrating nephroprotective characteristics. The highest levels of luteolin, chrysoeriol, and kaempferol were found in the plant stem (21.09, 45.19, and 27.55 mg g^{-1}, respectively), while the highest levels of quercetin, apigenin, and rutin were found in the flowers (20.10, 47.02, and 25.36 mg g^{-1}, respectively), and the highest levels of avicularin were found in the SDW (4.15 mg g^{-1}). Quercetin is a plentiful polyphenolic flavonoid that has a number of health-promoting properties, including strong vasodilators, cancer-preventative properties, anti-inflammatory properties, and advantages for asthma, among many others [55]. Additionally, luteolin and apigenin can lessen the occurrence of mouth sores and provide modest symptom relief as well as limit the viability of leukemic cells, colon and ovarian carcinoma cells, and, in particular, human breast cancer cells [56]. Moreover, kaempferol and avicularin are nontoxic and have potent antioxidant, hepatoprotective, antidiabetic, and anti-inflammatory properties [57].

Five phenolic substances were produced by the HPLC of the extracts of the different TR organs: ellagic acid, catechol, resorcinol, gallic acid, and phloroglucinol (Table A1 and Figure 2). The five compounds were found in plant stems and leaves, while resorcinol and gallic acid and catechol were not found in the SDW or flowers. The greatest content of gallic and ellagic acids (23.53 and 14.90 mg g^{-1}) in TR stems was lower than 37.4 and 50.3 mg g^{-1} recorded in the stem pruning wastes and flower residue recorded by Galal et al. [1] and Marlene et al. [58], respectively, in the same species. Numerous biological functions of gallic acid include antioxidant capabilities, antibacterial activity, anti-inflammatory properties, antiviral and antimutagenic characteristics, and anticancer effects [59]. Additionally, ellagic acid is a crucial compound utilized in the treatment of chronic ulcerative colitis as an anticarcinogenic, multipurpose protector against oxidative stress, and an anti-inflammatory agent [5]. Furthermore, catechol, resorcinol, and phloroglucinol concentrations were highest in the plant leaves (13.70, 11.91, and 6.24 mg g^{-1}, respectively). These compounds were discovered to have possible antimicrobial properties [10].

Alkaloids are physiologically-active substances that are frequently employed as medications and produced by plants as secondary metabolites, but many of these substances are extremely toxic [60]. Six alkaloid compounds were isolated and identified in TR extracts as berbamine, jatrorrhizine, palmatine, reticuline, isocorydine, and boldine (Table A1 and Figure 3). These compounds are widespread in many Chinese medicinal plants and have been shown to have leukocytosis-promoting, antimicrobial, anti-inflammatory, anticancer, and choleretic activities [61]. Galal et al. [1] found comparable results in the same species' pruning leaves and stem. Berbamine, jatrorrhizine, palmatine, and reticuline were all found in the highest concentrations (5.28, 5.83, 3.50, and 8.50 mg g^{-1}, respectively) in the plant stem, whereas isocorydine and boldine were found in the highest concentrations (9.11 and 4.79 mg g^{-1}, respectively) in the flowers. Berbamine, jatrorrhizine, palmatine, and reticuline have antibacterial activity, encourage leukocytosis, and are choleretic substances [62]. However, the long-term use of boldine may cause depression, partial motor aphasia, sound hallucinations, and color hallucinations [63].

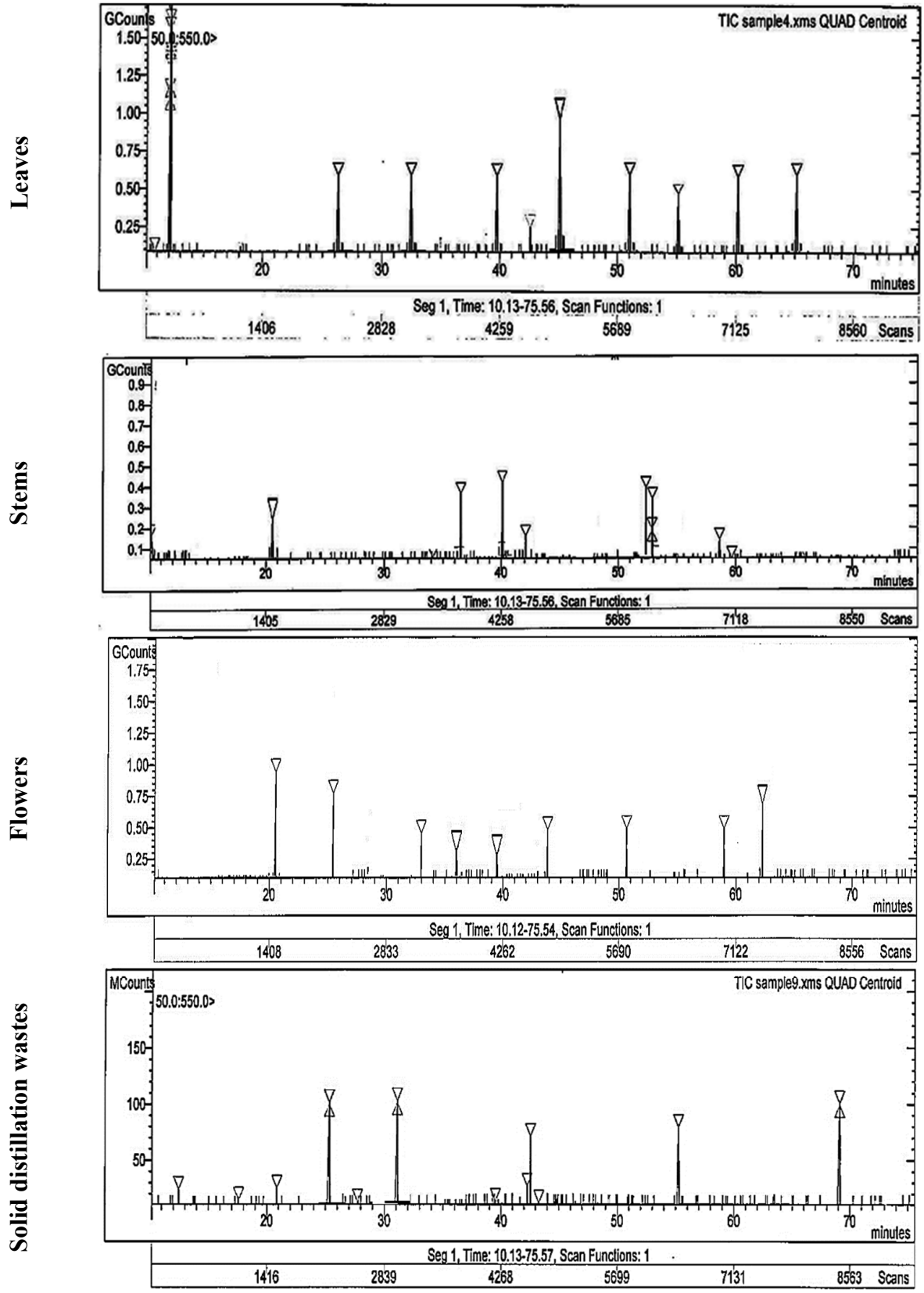


Figure 1. HPLC analysis of the phenolic compounds in the different organs as well as the solid distillation wastes of Taif's rose collected from Al-Shafa highlands in Taif City, Saudi Arabia.

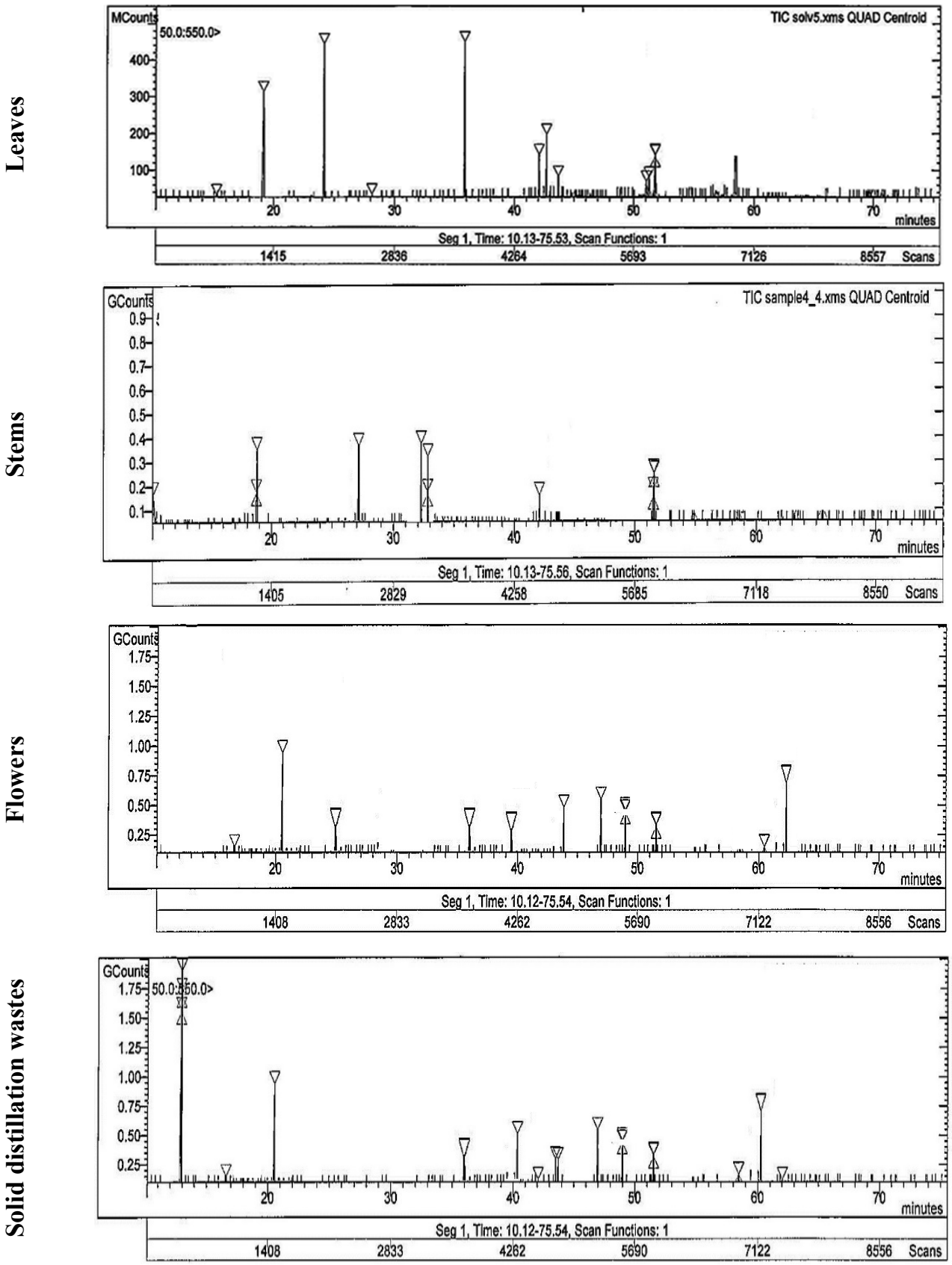


Figure 2. HPLC analysis of the flavonoid compounds in the different organs as well as the solid distillation wastes of Taif's rose collected from Al-Shafa highlands in Taif City, Saudi Arabia.

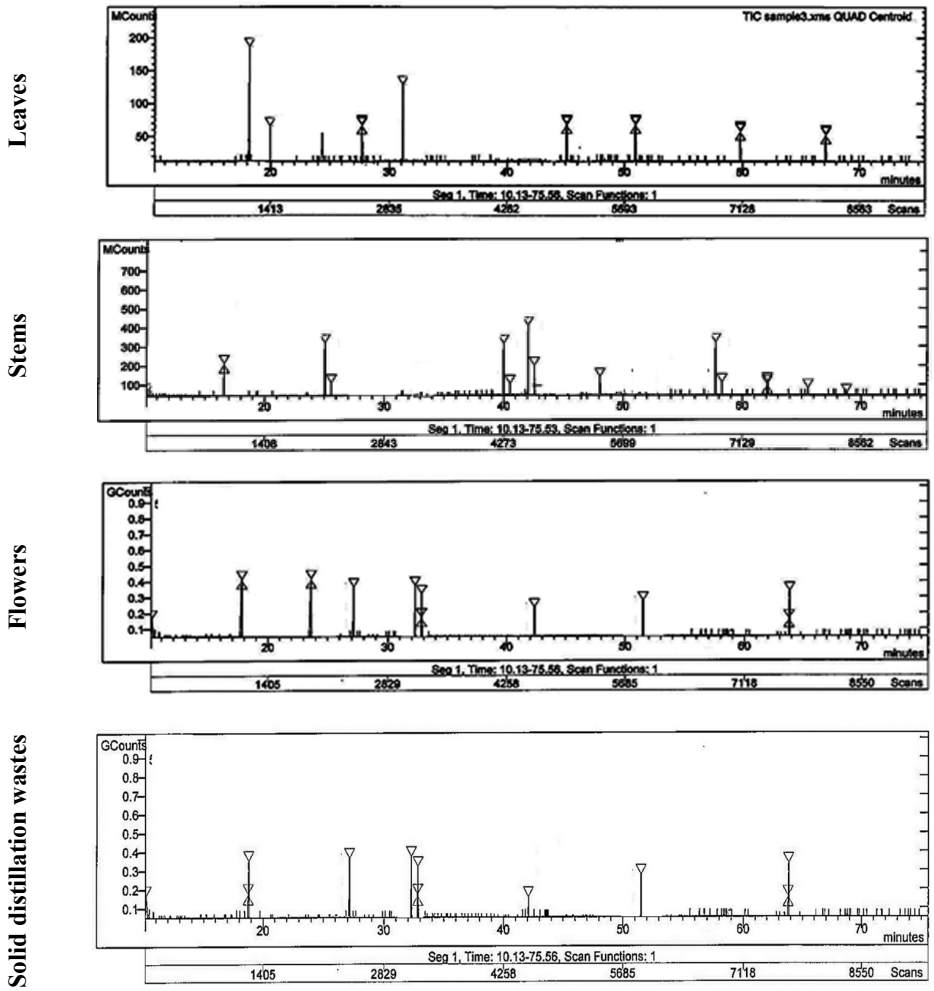


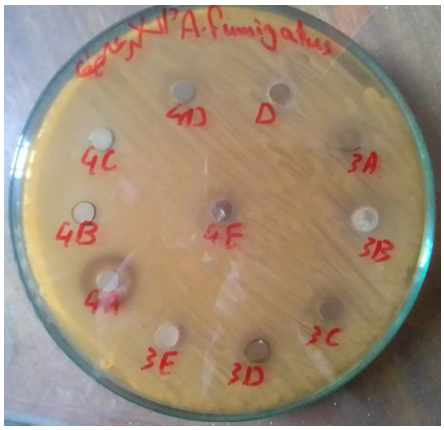
Figure 3. HPLC analysis of the alkaloid compounds in the different organs as well as the solid distillation wastes of Taif's rose collected from Al-Shafa highlands in Taif City, Saudi Arabia.

3.3. Biological Activity

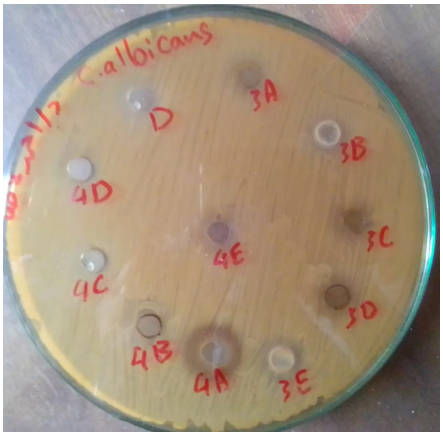
3.3.1. Flower Extracts

The phytogeographical region, the plant component, and the extraction method all affect a plant's antibacterial activity [64]. According to biological activity tests, TR flower extracts were effective against Gram-negative and Gram-positive bacteria but ineffective against fungal strains (Table A2 and Figure 4). In a related investigation, Galal et al. [1] discovered that the leaf pruning wastes of TR showed antibacterial and antifungal activity in the boiling-water extracts but not in the other extracts. Compared to the control (inhibition zones: 26 mm), ethanol and warm-water extracts significantly reduced the sensitivity of *Bacillus subtilis* (14 and 13 mm, respectively). Additionally, *Proteus vulgaris* and *Escherichia coli* were extremely sensitive to most extracts (except for warm water on *E. coli*), with boiling-water extracts having the maximum activity (13 and 18 mm, respectively) when compared to gentamicin (30 and 17 mm, respectively). In comparison to gentamicin antibiotics (control), Galal et al. [1] found that the same bacterial strains were much more sensitive to warm-water extracts of TR leaves and stem pruning wastes. They attributed these findings to the presence of various phytochemical compounds, such as alkaloids, phenolic acids, flavonoids, and other phenolic compounds. Similar findings on *Rosa indica* extracts show antifungal action against *A. fumigatus* strains as well as antibacterial activity against *Proteus* sp. and *E. coli* [65]. The current findings show that the floral extracts had a modest effect on the bacterial strains under investigation, with boiling- and cold-water extracts being the most effective against *P. vulgaris*. It is important to note that *P. vulgaris* was only responsive to TR extracts in the following order:

ethanol > warm water > methanol > boiling and cold water. Norziah et al. [66] claim that because it is nontoxic and beneficial to the environment, using water as an extracting solvent is preferable to using organic solvents. Additionally, water is a good solvent for extracting a significant amount of highly active phenolic and flavonoid chemicals that can be used safely in a variety of food applications [10].



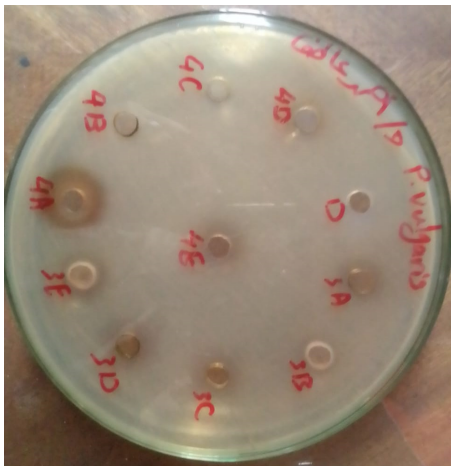
Aspergillus fumigatus



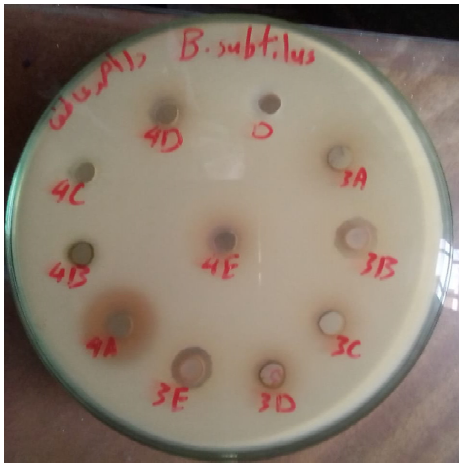
Candida albicans



Escherichia coli



Proteus vulgaris



Bacillus subtilis

Figure 4. Antimicrobial activity of the different extracts of Taif's rose. 3: flowers, 4: solid distillation wastes, A: methanol extract, B: ethanol extract, C: boiled water, D: cold water, E: warm water.

3.3.2. Solid Distillation Waste Extracts

The antimicrobial activity data of the SDW extracts of TR showed that the methanol extract was exclusively potent against fungal strains (*Aspergillus fumigatus* and *Candida albicans*) with lower activity (10 and 14 mm, respectively) compared with 17 and 20 mm, respectively, of the ketoconazole (Table A3 and Figure 4). Similarly, the methanol and cold-water extracts of the stem pruning wastes of TR were active against the bacterial and fungal strains [1]. On the other hand, the warm-water extract was only efficient against the investigated bacterial strains with inhibition zones (10, 12, and 18 mm) compared with 26, 30, and 17 mm for gentamicin. It was noticed that the SDW extracts had low efficacy against the investigated fungal and bacterial strains, except for the warmwater extract against *P. vulgaris*. Halawani [23] found that *P. vulgaris* was highly susceptible to all of the TR flower extracts. In contrast, Adom et al. [56] found that the aqueous extract of *P. major* has no antimicrobial activity. Thus, pharmaceutical studies are required to separate, purify, and identify the phytochemical compounds in the ethanolic, methanolic, and water extracts of TR flowers and SDW.

3.4. Nutritional Properties

3.4.1. Mineral Nutrients

Using organic fertilizer made from composted agricultural waste can help reduce the need for chemical fertilizers and nutrient requirements [67]. The statistical analysis (ANOVA I) indicated significant variations ($p < 0.05$) in the concentrations of the analyzed mineral elements (except Ca and Mg) among the different organs of TR (Table 2). The plant stem contributed the highest contents of N, K, and Mg (138.73 ± 16.49, 174.57 ± 17.20, and 96.12 ± 17.01 mg kg^{-1}, respectively), but the lowest Na content (136.15 ± 15.17 mg kg^{-1}). In addition, the plant leaves had the highest contents of P and Ca (6.58 ± 0.54 and 173.93 ± 26.03 mg kg^{-1}, respectively) but the lowest of K and Mg (101.54 ± 10.89 and 85.43 ± 14.90 mg kg^{-1}, respectively). The values of these elements (except K, P, and Ca) were lower than those recorded in the leaf and stem pruning wastes of the same species [17]. Moreover, the highest Na content (198.08 ± 12.34 mg kg^{-1}) was recorded in the flower tissues, which was comparable to 195.18 ± 9.32 mg kg^{-1} recorded in the SDW. The nutritional value of TR plant parts was lower than that found in herbal plant residues and olive mill waste ([68,69], respectively). The current study is in line with the findings of Galal et al. [17], who found that the macronutrient content of TR leaf and stem pruning wastes makes them more promising for composting in order to improve soil quality than flowers and SDW. The various plant organs of TR can be used as additives in organic fertilizers because their nutrient release rate is too slow to meet crop requirements in a timely manner. Based on the observation of Chang et al. [67], those organic fertilizers could not be an absolute replacement for chemical fertilizers.

Table 2. Mineral nutrient content (mean ± SD, n = 12) of the different organs of Taif's rose grown on Al-Shafa highlands in Taif City, Saudi Arabia. Maximum and minimum values are underlined.

Organ	N	P	K	Ca	Mg	Na
	mg kg^{-1}					
Leaves	98.01 ± 3.86 ab	6.58 ± 0.54 a	101.54 ± 10.89 b	173.93 ± 26.03	85.43 ± 14.90	167.64 ± 19.09 ab
Stem	138.73 ± 16.49 a	6.53 ± 1.77 a	174.57 ± 17.20 a	165.46 ± 24.28	96.12 ± 17.01	136.15 ± 15.17 b
Flower	71.70 ± 5.31 b	1.49 ± 0.19 b	117.10 ± 2.01 ab	147.54 ± 17.20	91.32 ± 12.01	198.08 ± 12.34 a
Distillation wastes	81.58 ± 6.23 b	2.24 ± 0.17 b	102.13 ± 13.11 b	135.55 ± 13.93	89.83 ± 11.73	195.18 ± 9.32 a
F-value	4.55 *	24.83 ***	5.5 **	0.96	0.22	4.01 *

Means in the same column followed by different letters are significantly different at $p < 0.05$, according to Duncan's HSD test; *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$.

3.4.2. Organic Nutrients

Animal productivity depends on forage quality, which is primarily determined by total protein (TP), crude fiber (CF), digestibility, and other related factors [70]. The statistical

analysis (ANOVA I) of the organic nutrients (except CF and NDF) showed significant variation ($p < 0.05$) among the investigated samples of TR plants (Table 3). The plant leaves contributed the highest percentage of total carbohydrates (NFE) and ADL (59.85 and 3.93%, respectively) but the lowest CF content (21.86%). The NFEs of TR leaves, stems, and flowers were comparable to those recorded in the pruning wastes of the same species [17], *Echinochloa stagnina* and *Eichhornia crassipes*, and higher than that of *C. demersum* (33.4%) [71]. The high carbohydrate content is beneficial for giving the rumen microorganisms sufficient energy, which in turn helps lactating cows [72]. Moreover, the highest percentages of TP and ADF (8.66 and 24.17%, respectively) were recorded in the plant stem associated with the lowest ash and fat contents (7.34 and 0.14%). The ADF content of the different investigated organs was lower than that of TR pruning wastes (27.5–45.1%: Galal et al. [17]), *Trifolium alexandrinum* (32.1%: [73]), and wheat straw (46.5–50.8%: [74]). The percentages of NDF and ADF of TR organs were lower than 74.4% and 44.4%, respectively, which were recorded in central Oklahoma [75] and 57.5% and 31.6%, respectively, reported in western Washington [76] for forage intermediate wheatgrass. Favre et al. [77] reported 59.7% NDF and 33.7% ADF in Wisconsin's Kernza intermediate wheatgrass in the similar context.

Table 3. Organic nutrient content (mean ± SD, $n = 12$) of the different organs of Taif's rose grown on Al-Shafa highlands in Taif City, Saudi Arabia. Maximum and minimum values are underlined.

Organ	Total Protein (%)	NFE (%)	Ash (%)	Fat (%)	Crude Fiber (%)	ADF (%)	ADL (%)	NDF (%)
Leaves	6.13 ± 1.69 ab	59.85 ± 9.96 a	11.67 ± 2.69 c	0.50 ± 0.05 ab	21.86 ± 3.48	22.88 ± 7.11 a	3.93 ± 1.12 a	38.21 ± 7.88
Stem	8.66 ± 2.27 a	52.63 ± 13.81 a	7.34 ± 1.75 d	0.14 ± 0.01 c	31.33 ± 3.81	24.17 ± 5.16 a	2.49 ± 0.49 ab	36.51 ± 3.31
Flower	4.71 ± 0.49 b	53.92 ± 5.15 a	18.74 ± 2.34 b	0.36 ± 0.05 b	22.26 ± 2.29	15.54 ± 1.41 ab	1.71 0.09 b	38.51 ± 3.36
Distillation wastes	5.13 ± 0.38 b	28.39 ± 4.78 b	29.39 ± 2.87 a	0.57 ± 0.05 a	36.52 ± 1.58	14.23 ± 1.75 b	3.60 ± 0.31 a	34.56 ± 1.53
F-value	4.31 *	5.28 **	62.75 ***	14.93 ***	2.71	2.96 *	2.85 *	0.46

Means in the same column followed by different letters are significantly different at $p < 0.05$, according to Duncan's HSD test; *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$.

Moreover, the flower SDW with the lowest values of NFE, ADF, and NDF (28.39, 14.23, and 34.56%, respectively) had the highest ash, lipids, and CF contents (29.39, 0.57, and 36.52%, respectively). According to Heneidy and Halmy [78], TP and CF are traditionally used as indicators of the nutritional content of food for grazing animals because TP is used for energy and aids in tissue formation, and CF defines the energy feeding value of the forage [79]. The minimal protein in animal diets ranges from 6 to 12% DM MAFF [80] and, as a result, the TP content of TR's leaves and stems falls within this range but that of the flowers and SDW is lower. In addition, the plant leaves and stems lie within the required range (7–9%) of TP for sheep and gestating cows ([45,81], respectively). CF in plants represents all the cell wall fractions that are resistant to the action of digestive enzymes and includes the insoluble residue of acid hydrolysis and the alkaline one [82]. In the current study, the CF contents of the SDW and stem of TR were slightly higher than the range reported for some known wild forage plants such as *Phragmites australis* (29.9% DM), *Panicum repens* (27.3% DM), and *Cynodon dactylon* (20.5% DM) [83,84]. However, the range of CF contents in the investigated organs of TR was higher than the mean content (20.0%) of temperate grasses [85].

3.4.3. Nutritional Value

The nutritive value of any forage is dependent upon its content of energy-producing nutrients as well as its contents of essential nutrients to the body and mainly depends on high digestible crude protein (DCP) [71]. The investigated parameters of nutritional values (except NE) were significantly varied ($p < 0.05$) among the different organs of TR plants (Table 4). DCP is an important fraction of proteins that are ingested and absorbed by the animal and not excreted in feces [86]. The highest values of DCP and GE (4.52% and 412.61 Mcal kg^{-1}, respectively) and the lowest TDN (56.18%) were recorded in the plant stem, while the highest TDN (59.19%) associated with the lowest DCP (0.86%) were recorded in the plant flowers. The DCP in the present study was relatively low

in comparison with TR pruning wastes [17] but was lower than 9% of the main fodder crop *T. alexandrinum* [87]. Additionally, the TDN is defined as the energy content of feeds available to animals after the digestion losses [79]. The present investigation revealed that the different organs as well as the SDW of TR have suitable contents of TDN for mature dry gestating beef cows that require 55–60% [88]. Moreover, the plant leaves had the highest values of DE, ME, and NE (2.68, 2.20, and 1.10 Mcal kg^{-1}, respectively). The SDW had the lowest energy contents represented by DE, ME, NE, and GE (1.23, 1.23, 0.62, and 324.11 Mcal kg^{-1}, respectively). The values of DE and ME of the different living organs of TR were comparable to the 2.65 and 2.17 Mcal kg^{-1} DM values, respectively, recorded in hay of alfalfa (*Medicago sativa*), and higher than the 2.43 and 1.99 Mcal kg^{-1} DM values, respectively, in red clover (*Trifolium pratense*) [45]. However, the values of the GE of the TR stems and leaves were comparable to the 377.02–424.25 Mcal kg^{-1} DM of the pruning wastes [17] and higher than the 389 Mcal kg^{-1} DM reported for *Cynodon dactylon* and 398 Mcal kg^{-1} DM in *Panicum repens* [84].

Table 4. Nutritional value (mean ± SD, $n = 12$) of the different organs of Taif's rose grown on Al-Shafa highlands in Taif City, Saudi Arabia. Maximum and minimum values are underlined.

Organ	DCP (%)	TDN (%)	DE (Mcal/kg)	ME (Mcal/kg)	NE (Mcal/kg)	GE (Mcal/kg)
Leaves	2.71 ± 0.44 ab	58.28 ± 1.87 ab	2.68 ± 0.37 a	2.20 ± 0.31 a	1.10 ± 0.01 a	385.58 ± 8.24 b
Stem	4.52 ± 0.16 a	56.18 ± 1.82 b	2.61 ± 0.42 a	2.14 ± 0.42 a	1.07 ± 0.03 a	412.61 ± 9.59 a
Flower	0.86 ± 0.05 b	59.19 ± 3.13 a	2.25 ± 0.19 a	1.85 ± 0.16 a	0.93 ± 0.06 a	354.33 ± 6.62 c
Distillation wastes	1.24 ± 0.35 b	59.05 ± 2.32 a	1.23 ± 0.08 b	1.23 ± 0.08 b	0.62 ± 0.04 b	324.11 ± 9.13 d
F-value	4.31 *	5.28 **	62.75 ***	14.93 ***	2.71	2.96 *

Means in the same column followed by different letters are significantly different at $p < 0.05$, according to Duncan's HSD test; *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$.

4. Conclusions

Compared to the flowers and SDW, the leaves and stems of TR contained more carbohydrates and cardiac glycosides but fewer flavonoids and phenolic substances. The largest cardiac glycoside, flavonoid, and phenolic levels were found in the TR flowers, with no discernible difference from the SDW, whereas the highest carbohydrate and alkaloid contents were found in the TR leaves. Quercetin, apigenin, and rutin flavonoids, as well as isocorydine and boldine alkaloids, were found in larger concentrations in the flowers and blossom SDW of TR plants than in the leaves and stems, according to the HPLC analysis of the phytochemical components. The various TR flower extracts were effective against Gram-negative and Gram-positive bacteria but had no effect on fungal strains, while the SDW's methanol extract was exclusively effective against fungi. In contrast to the flowers and SDW, the macronutrient content of TR leaves and stems makes it possible to compost them for improving soil quality. The results of the current investigation show that the TDN contents of the various organs and the SDW of TR are suitable for mature dry gestating beef cows. In addition to the potential use of the SDW as a supplement in animal feed, the various plant organs and the SDW of TR can be utilized for a variety of medicinal purposes. Based on these micro-components as well as the inorganic and organic nutrients and the nutritive value, the different organs of TR can be used in medicine, animal fodder, soil amendment, organic fertilizers, and for other industrial purposes (Table 5).

Table 5. Micro-components of the different organs of Taif's rose and their significance.

Component	Significance
Inorganic nutrients (N, P, Na, K, Ca, Mg)	Soil amendments, organic fertilizers
Organic nutrients (carbohydrates, proteins, fats, fibers)	Animal forage, industrial application
Nutritional components (DCP, TDN, GE, ME, NE)	Animal forage
Secondary metabolites (cardiac glycosides, flavonoids, alkaloids, phenolics)	Medicinal purposes (e.g., cancer-preventative properties, antioxidant, anti-inflammatory, and antimicrobial), pharmaceutical industry.

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Conflicts of Interest: The authors declare no conflict of interest.

Appendix A

Table A1. HPLC analysis of the phenolic, flavonoid, and alkaloid contents of the different organs as well as the solid distillation wastes of Taif's rose collected from Al-Shafa highlands in Taif City, Saudi Arabia. SDW: solid distillation wastes; ND: not detected.

Chemical Compound		Plant Organ			
		Leaves	Stem	Flowers	SDW
Flavonoids concentration ($mg\ g^{-1}$)	Quercetin	11.09	12.61	20.10	2.69
	Apigenin	4.59	23.83	47.02	11.25
	Luteolin	10.17	21.09	13.60	ND
	Chrysoeriol	19.20	45.19	ND	6.98
	Rutin	16.87	11.16	25.36	ND
	Kaempferol	21.59	27.55	19.10	9.68
	Avicularin	ND	ND	2.50	4.15
Phenolics concentration ($mg\ g^{-1}$)	Gallic acid	14.21	23.53	5.41	ND
	Ellagic acid	7.94	14.90	11.69	7.25
	Catechol	13.70	6.59	8.60	ND
	Resorcinol	11.91	7.57	ND	8.79
	Phloroglucinol	6.24	0.93	1.69	3.45
Alkaloids concentration ($mg\ g^{-1}$)	berbamine	3.03	5.28	ND	ND
	jatrorrhizine	5.10	5.83	5.60	4.26
	palmatine	ND	3.50	ND	3.44
	reticuline	1.88	8.50	5.30	ND
	isocorydine	1.60	4.70	9.11	4.77
	boldine	ND	0.64	4.79	3.56

Table A2. Antimicrobial activity (mm) of the different extracts of Taif's rose flowers on the pathogenic fungal and bacterial strains. NA: no activity.

Extract	Fungi		Bacteria		
			Gram- + ve Bacteria	Gram- — ve Bacteria	
	Aspergillus fumigatus	*Candida albicans*	*Bacillus subtilis*	*Escherichia coli*	*Proteus vulgaris*
Control	Ketoconazole		Gentamicin	Gentamicin	
	17	20	26	30	17
Methanol	NA	NA	NA	10	16
Ethanol	NA	NA	14	11	13
Boiling water	NA	NA	NA	13	18
Cold water	NA	NA	NA	12	18
Warm water	NA	NA	13	NA	14

Table A3. Antimicrobial activity (mm) of the different extracts of Taif's rose solid distillation wastes on the pathogenic fungal and bacterial strains. NA: no activity.

Extract	Fungi		Bacteria		
			Gram- + ve Bacteria	Gram- — ve Bacteria	
	Aspergillus fumigatus	*Candida albicans*	*Bacillus subtilis*	*Escherichia coli*	*Proteus vulgaris*
Control	Ketoconazole		Gentamicin	Gentamicin	
	17	20	26	30	17
Methanol	10	14	10	19	NA
Ethanol	NA	NA	NA	NA	NA
Boiling water	NA	NA	NA	NA	NA
Cold water	NA	NA	NA	14	12
Warm water	NA	NA	10	12	18

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