ISSN (Online): 2091-2609
DOI Prefix: 10.3126/ijasbt

International Journal of Applied Sciences and Biotechnology (IJASBT)

Research Article

Unveiling The Potential of Jimbu *(Allium Przewalskianum)*: Bioactive Compounds, Antioxidant, and Antimicrobial Properties of a Native Himalayan Spice Herb

Shreejana Pandey[1], Guheswari Chataut[1]*, Suja Maharjan[1], Dev Raj Maidali[1], Keshab Bhattarai[2]

[1]Central Department of Biotechnology, Tribhuvan University, Kathmandu, Nepal.
[2]Sattwik Nutrifood A diet therapy and Research Center, Sattwik Group Pvt Ltd, Pokhara, Nepal.

Article Information

Received: 16 October 2023.
Revised version received: 14 December2023.
Accepted: 17 December 2023.
Published: 29 December 2023.

Cite this article as:
S. Pandey et al. (2023) Int. J. Appl. Sci. Biotechnol. Vol 11(4): 171-180. DOI: 10.3126/ijasbt.v11i4.59326

***Corresponding author**
Guheswari Chataut,
Central Department of Biotechnology, Tribhuvan University, Kathmandu, Nepal
Email: chatautguheswari@gmail.com

Peer reviewed under authority of IJASBT

Abstract

Jimbu (*Allium przewalskianum*), is an indigenous Himalayan spice herb in Nepal and has a long history of culinary use, yet its potential beyond home kitchens remains largely unexplored due to limited studies and exposure. It does not require extensive input of cash, machinery, land, or labor to profit from, making it an excellent and valuable source of pharmaceutical ingredients owing to its bioactive compounds. We investigated its antioxidant and antimicrobial properties. Quantitative analysis of Jimbu extracts revealed the presence of bioactive compounds, including Oleoresin (extracted using acetone and methanol), polyphenols, flavonoids, and tannins. The methanol extract of. *A.* przewalskianum exhibited the highest polyphenol content (8.11±0.24 mg GAE/g), flavonoid content (5.28±0.29 mg GAE/g) and tannin content (0.923±0.02 mg GAE/g). The antioxidant potential of Jimbu was seen to be increased with an increase in concentration, reaching 58.79% at 400µg/ml, with an IC50 value of 303.58µg/ml for DPPH radical scavenging activity. Likewise, the yield of oleoresin was obtained as (10.86±0.27) from acetone and (7.8±0.20) from methanol. Similarly, sensory evaluation through One-way ANOVA highlighted concentrations of 0.07% and 0.1% of Oleoresin to be most favored. Furthermore, sensory evaluation of the Jimbu extracts in minced chicken meat exhibited remarkable antimicrobial activity against Salmonella along with a significant reduction in the colony forming units (CFU). This study shows that the extracts of *A. przewalskianum* could be utilized as a potential medicinal plant for the development of effective drugs against pathogenic bacteria. Meanwhile, it provides opportunities for economic benefits and a new potential revenue source for rural communities in the Himalayan region of Nepal.

Keywords: Jimbu, Oleoresin, Antioxidant, Bioactive compounds, Antimicrobial activity.

Introduction

Throughout history, people have utilized plants for both sustenance and medicinal purposes, reflecting a profound and enduring connection between humans and their environment (Nepal, 2006). Traditional herbal remedies have been developed over centuries to prevent and treat various ailments, including degenerative diseases such as cancer and cardiovascular disorders. In many developing countries, traditional herbal medicine remains a primary healthcare resource for the majority of the population. Phytochemicals found in plants, including phenolics, carotenoids, anthocyanins, and tocopherols have shown potential in preventing chronic diseases and protecting the body from oxidative damage caused by free radicals (Abu-

Bakar *et al*., 2009). Phenolic acids and flavonoids, in particular, are potent antioxidants with reported antibacterial, antiviral, anticarcinogenic, anti-inflammatory, and vasodilatory properties (Galeotti *et al*., 2008).

Oxidative reactions in food limit their shelf life and contribute to various diseases. Increased use of polyunsaturated fatty acids and light-weight, oxygen-permeable packaging in pursuit of healthier and more sustainable food options has heightened concerns about oxidative rancidity (Huang and Ahn, 2019). Synthetic antioxidants like butylated hydroxytoluene (BHT) and butylated hydroxyanisole (BHA) have been widely employed to mitigate lipid oxidation. With growing safety concerns and the preference for natural products, there is a rising demand for natural antioxidants (Kapadiya *et al*., 2016).

Manufacturers of food, beverages, cosmetics, and pharmaceuticals are responding to consumer resistance and regulatory restrictions on chemical additives. Similarly, the contamination of poultry meat with foodborne pathogens, including *Salmonella*, remains a public health concern due to potential illness resulting from improper handling, cooking, or storage (Rouger *et al.*, 2017). Thus, spices and herbs have emerged as excellent sources for food preservation, offering a healthier and more consumer-friendly alternative to synthetic antioxidants (Kapadiya *et al*., 2016).

In addition to this, phenolics, essential oils, and oleoresins found in spices have exhibited significant antimicrobial and antioxidant properties. (Diniz do Nascimento *et al*., 2020). Oleoresins are the concentrated liquid form of the spice. Spice oleoresins represent the complete flavour profile of the spice. It contains the volatile as well as non-volatile constituents of spices (Unila, 2015).

The major spices grown in Nepal are Akhbare khursani (*Capsicum annum*), Dalchini (*cinnamon*), Black salt, Black *cumin*, Alaichi (*Amomum sabulatum*), Jwano (*Trachyspermum amni*), Coriander, Methi (*Trigonella foenum-graecum*), Jeera (*cumin*), Turmeric (*Curcuma longa*) and Timmur (*Sichuan pepper*). Among these, Jimbu stands out as a native product of the north-central mountainous region of Nepal, particularly valued in Upper Mustang. Jimbu, derived from two *Allium* species, *A. hypsistum* and *A. przewalskianum*, both belonging to the Amaryllidaceous family (Shrestha *et al*., 2000), is a versatile spice used for flavoring lentil dishes, stir-fried vegetables, salads, pickles, and more. Remarkably, 46% of households in Upper Mustang employ Jimbu as traditional medicine to treat human and livestock ailments, particularly coughs, colds, high-altitude sickness, and stomach pains (IUCN Nepal, 2000). Additionally, Jimbu significantly contributes (10%) to the annual income of households in this region. It can be considered as a highly valued cash crop with enormous capacity of international trade that can enhance incomes and livelihoods (Nepal, 2006). The dried Jimbu is generally stored in a dry, low-humidity, enclosed area after it is harvested, and meaning there would be no need to refrigerate the spice when exporting it. Jimbu does not require a Phytosanitary Certificate or a Permit to Import, and like other spices would be regarded as a low-risk import product (CFIA, 2014).

Materials and Methods

Plant Materials, Sample Preparation and Extraction

Jimbu was purchased in April 2017, from the local market Aasan Kathmandu, Nepal and mechanically reduced to powder form using a grinder. The powder was then sifted through a laboratory sieve of a 60-mesh size (0.0098 inch). The grits were removed and the powder was packed in plastic pouch. To extract the desired compounds, 5-10 g of each sample was mixed with 80% methanol (30 ml) and agitated continuously for 20 minutes. The resulting mixture was then filtered through Whatman no. 1 filter paper. The residue was again submitted to two more extraction cycles of 20 minutes each, resulting in a total extraction time of 60 minutes. The filtrate was combined in a volumetric flask, and the volume was adjusted to 100 ml (Sultana *et al*., 2008). The extracts were stored in the refrigerator until analysis.

Phytochemical Screening

Major Phytochemicals present in the extracts of Jimbu were screened by using standard qualitative methods. Assays were carried out to ascertain flavonoids, phenolics and tannins.

Determination of Total Phenol Content

The quantification of total polyphenol content was conducted by Folin-Ciocalteau colorimetry, described in a study by Samarth *et al*., 2008. 1 ml of extract was poured into a 25 ml volumetric flask containing 9 ml of distilled water. To this mixture, 1 ml of Folin-Ciocalteu reagent was added and mixed thoroughly. After incubation of about 5 minutes, 10 ml of 7% sodium bicarbonate (Na_2CO_3) solution was added followed by incubation of 90 minutes at room temperature. Finally, the absorbance was measured at a wavelength of 765 nm. Total polyphenol content was reported as mg GAE/g. The calibration curve was constructed to determine the corresponding Gallic acid concentration in the samples.

Determination of Total Flavonoid Content

The total flavonoid content (TFC) was determined as per (Samatha *et al*., 2012) using aluminium chloride assay through colorimetry. An aliquot (0.5 ml) of extract was taken in different test tubes and 2 ml of distilled water was added followed by the addition of 0.15 ml sodium nitrite (5% $NaNO_2$, w/v) and was allowed to stand for 6 minutes. Later 0.15 ml of aluminium trichloride (10% $AlCl_3$) was

added and incubated for 6 minutes, followed by the addition of 2 ml of sodium hydroxide (NaOH, 4% w/v) and volume was made up to 5 ml with distilled water. The mixture was then incubated for 15 minutes and absorbance at 510 nm was taken**.** The standard calibration curve was prepared by preparing gallic acid solutions and results were expressed as mg of Gallic acid equivalents per litre of sample.

Determination of Total Tannin Content

The tannin was determined by the Folin-Ciocalteu method. For this, 0.1 ml of the sample extract was added to a volumetric flask (10 ml) containing 7.5 ml of distilled water. To this mixture about 0.5 ml of Folin-Ciocalteu phenol reagent was pipetted followed by the addition of 1 ml of 35 % Na_2CO_3 solution and then diluted to 10 ml using distilled water. After this, the mixture was shaken well and kept at room temperature for 30 minutes. Then, a set of reference standard solutions of Gallic acid (20, 40, 60, 80 and 100 µg/ml) were prepared. Eventually, absorbance for test and standard solutions was measured against the blank at 725 nm with a UV/Visible spectrophotometer. The tannin content was thus expressed in terms of mg GAE /g of extract.

Extraction of Oleoresin and Calculation of Its Yield Percentage

Oleoresin content of Jimbu was obtained using methanol and acetone as per AOAC and the yield percentage of oleoresin was calculated using the following formula and then stored at 4ºC.

$$\text{Yield percentage} = \frac{\text{Weight of oleoresin /EO obtained}}{\text{Weight of dried spice taken}} \times 100\%$$

Selection of Desired Concentration of Oleoresin

Minced chicken was fried to perform sensory evaluation. It was divided into 4 different parts each weighing about 25 g. Further, 200 mg salt was added to each sample for enhancement of the taste. To select the suitable concentration of oleoresin, all parts were treated with different concentrations of oleoresin (0.03%, 0.07% and 0.10%). The oleoresin concentrations under study were assigned as A (0.03%), B (0.07%) and C (0.10%) for easy analysis and clear depiction of obtained results. Then sensory analysis was carried out for each sample.

Sensory Analysis

Sensory analysis was performed using a 9-point hedonic rating (Peryam and Pilgrim, 1957). The sensory evaluation for overall quality was done with 15 semi-trained panelists from National College of Food Science and Technology, Kathmandu. The appearance, smell, color, flavor and overall acceptability were the parameters used for the sensory evaluation,

Sample Preparation

Chicken keema was collected from the local market of Naya bazaar, Kirtipur, Nepal. The selected concentrations of oleoresin, 0.07% and 0.1% were inoculated in the minced chicken meat. Then the samples were taken for microbial analysis immediately after inoculation (i.e., day 0) and again after 3, 5, and 7 days of refrigerated storage.

Total Viable Count

Total viable count (TVC) was determined on Plate Count Agar (PCA) following the standard method by AOAC, 2005. 25-250 colony- forming units (CFU) were counted and the results were expressed in logarithmic of colony-forming units per gram of meat (log CFU/g).

Enumeration of Salmonella

Salmonella that grows on xylose lysine deoxycholate agar (XLD) medium were isolated by serial dilution method followed by pour plate technique.

Determination of Total Antioxidant Activity

The antioxidant activity was determined by the DPPH radical scavenging method as described by Walvekar and Kaimal, 2014 and Alam *et al.*, 2013. 1 ml of extract containing 1 mg/ml of dry matter in methanol was mixed with 4 ml of deionized water and 1 ml of freshly prepared solution of DPPH in methanol (0.004% w/v). The mixture was shaken vigorously and left for 30 minutes in the dark room. And absorbance was taken at 517 nm. The antioxidant activity as DPPH inhibition % was calculated as %.

$$\text{Antioxidant activity as DPPH inhibition} = \frac{(A-B)}{A} \times 100$$

Where A is the absorbance of DPPH and B is the absorbance of DPPH and extract combination

Results and Discussion

Natural phytochemicals derived from fruits, vegetables and herbs have been reported to possess a wide range of biological effects, including antioxidant, antimicrobial and anti-inflammatory actions (Shin *et al.*, 2020). Oleoresin present in Jimbu can be used as a source of natural antimicrobials. Apart from this, spices like Jimbu are preferred because of their uniformity in flavour and pungency, ease of storage and transport (Unila, 2015). Diarrhoea or loose dung, cough and cold, and lung and liver diseases are the major livestock illnesses treated with Jimbu (IUCN Nepal, 2000).

Antioxidant Activity Analysis of Jimbu

The percent inhibition method was used to determine the antioxidant activity of Jimbu. The methanolic extract of Jimbu significantly and dose -dependently reduced DPPH. The antioxidant efficacy of Jimbu was found to have increased with an increase in the concentration of methanolic extract.

The antioxidant activity of *Allium przewalskianum* was found to be 58.79% at 400 µg/ml. The antioxidant activity of *Allium cepa* was shown to be 35% at 400 µg/ml by Kaur et al., 2016 which is lower than the antioxidant activity

exhibited by Jimbu in this study. The research on *Allium sativum* and *Allium cepa* also shows that % DPPH inhibition is dependent on concentration. The R square of this fitted model is 0.9323. This shows that 93.23% of the increase in % DPPH inhibition is due to the effect of concentration. There is a statistically significant association ($p < 0.05$) between concentration and % DPPH inhibition. DPPH free radical scavenging was observed to evaluate the antioxidant activity of Jimbu. The inhibitory concentration expressed as IC_{50} (half inhibitory concentration) was calculated from the data illustrated in Fig. 1 and was compared with IC_{50} value of standard ascorbic acid as shown in Fig. 2. At a concentration of 400 µg/ml, extract of the Jimbu scavenged over 58.79% of DPPH radicals with an IC_{50} value of 303.58µg/ml which was greater than the IC_{50} (129.68µg/ml) value of ascorbic acid.

Determination of TPC, TFC and TTC

The total phenolic content (TPC), total flavonoid content (TFC) and total tannin content (TTC) of the extract were expressed in terms of gallic acid equivalent (mg GAE/g) dry weight of the extract, calculated using standard gallic acid curve (Fig. 3). The TPC of the methanol extract and petroleum extract of Jimbu was found to be 8.11±0.24 and 6.94±0.52 mg GAE/g respectively. The methanol extract of a Jimbu showed the highest content of polyphenol. A study conducted by (Mamun et al., 2016) on *Allium sativum*, shows that the extractability of polyphenol content depends on the type of solvent used.

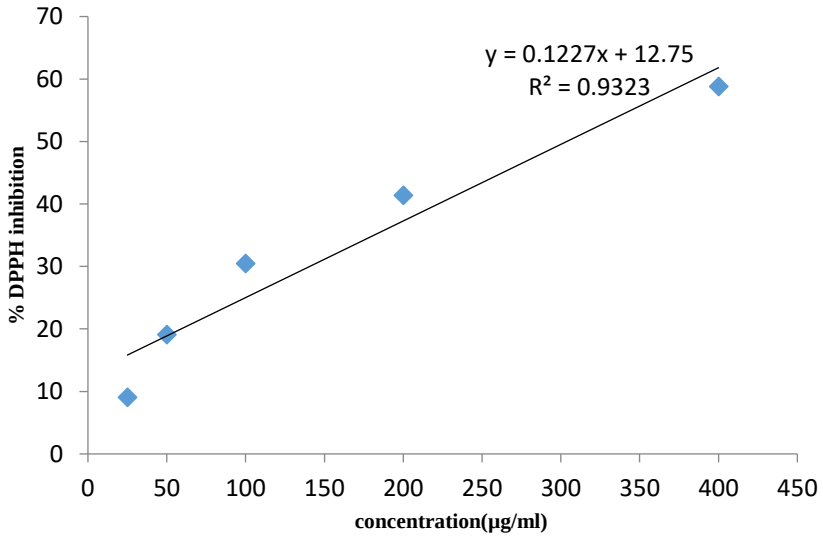

Fig. 1: Antioxidant activity as DPPH inhibition and concentration of *jimbu* extract

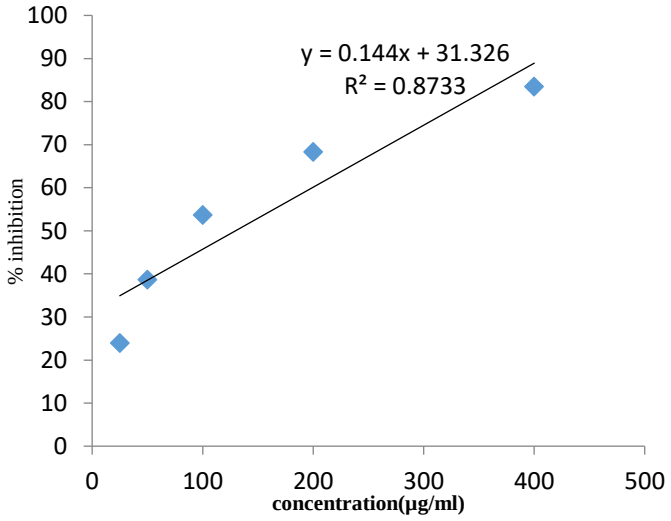

Fig. 2: Standard Ascorbic acid Equivalent curve at 517nm

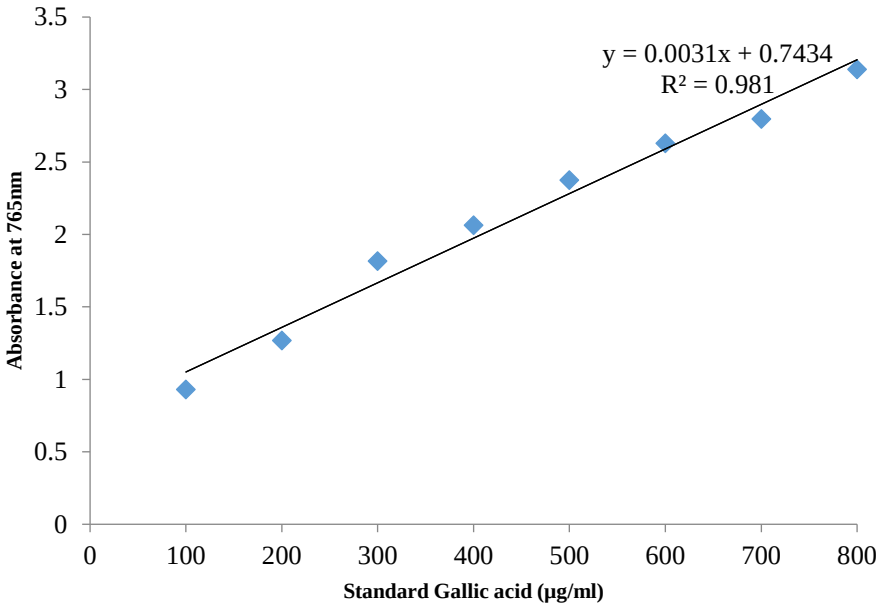

Fig. 3: Standard Gallic Acid Equivalent Calibration Curve for Polyphenol at 765nm

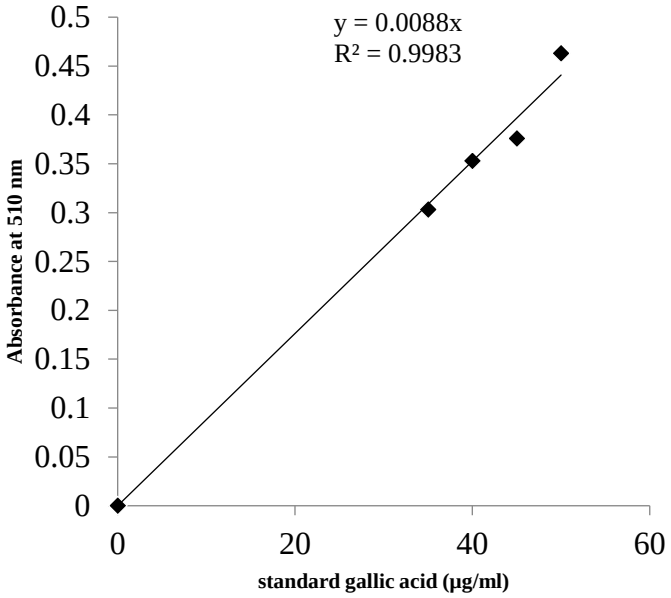

Fig. 4: Standard Gallic Acid Equivalent Curve for flavonoid at 510nm

The Fig. 4 shows flavonoid content of the methanol extract and petroleum ether extract of Jimbu was found to be 5.28±0.29 and 4.32±0.17 mg GAE/g respectively. The methanol extract of Jimbu showed the highest content of flavonoids. Results were found statistically significant ($p < 0.05$). The type of solvent used and flavonoid content are correlated. Lin *et al*., 2009, mentioned that methanol can dissolve phenolic compounds, tannin, flavonoid, saponin, and anthocyanin more effectively than petroleum ether.

The tannin content of the methanol extract and petroleum ether extract of Jimbu was found to be 0.92±0.02, 0.64±0.06 mg GAE/100g respectively. The methanol extract of Jimbu possessed the highest tannin content. Rajesh et al. (2016) also presented similar results (Fig. 5).

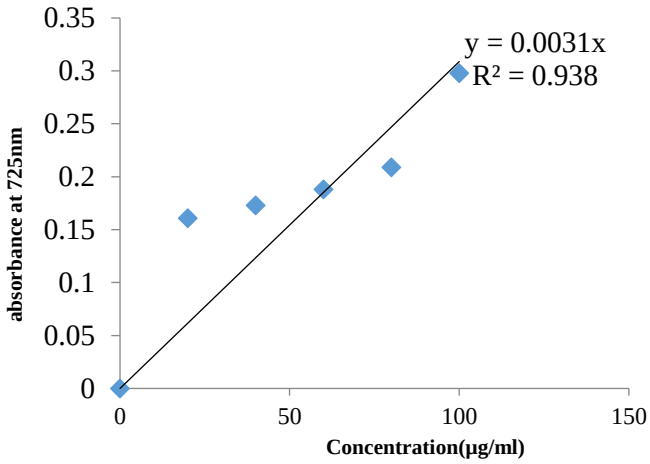

Fig. 5: Standard Gallic Acid Equivalent Curve for tannin at 725nm

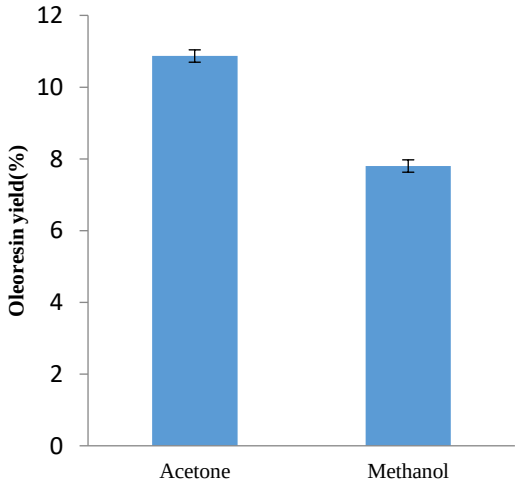

Fig. 6: Yield of Oleoresin using two different solvents

Effect of Solvents on Oleoresin Yield

The oleoresin content of Jimbu was obtained using two solvents. The yield of oleoresin from Acetone and Methanol was found to be 10.86±0.27, 7.8±0.20 respectively.

The highest oleoresin yield was obtained from acetone as shown in Fig. 6. The bar diagram above and table 1 showed that the results were statistically significant in oleoresin yield between acetone and methanol ($p < 0.05$). In simpler terms, the choice of solvent appears to have a notable impact on the extraction of oleoresin, and the difference observed is not likely due to random chance. The findings highlight the importance of selecting the right solvent for oleoresin extraction, as it significantly influences the outcome. Furthermore, these findings are supported by the reported results from the study conducted by (Rezazi et al., 2017), which mentions that acetone gives a higher oleoresin yield than methanol and oleoresin yield is affected by the nature of extracting solvent. Many factors contribute to the efficiency of solvent extraction, such as the type of solvent, the temperature, the number of steps and the particle size and shape of the plant matrix (Taher & Sarmidi, 2015).

Table 1: t-test for oleoresin yield

	Acetone	*Methanol*
Mean	10.86667	7.8
Variance	0.223333	0.13
Observations	3	3
Pooled Variance	0.176667	
Hypothesized Mean Difference	0	
Degrees of freedom	4	
t Stat	8.93583	
P(T<=t) one-tail	0.000434	
t Critical one-tail	2.131847	
P(T<=t) two-tail	0.000867	
t Critical two-tail	2.776445	

Sensory Evaluation Analysis

One-way ANOVA was used to understand the significant difference between all samples based on appearance, smell color, flavor and overall acceptability analyzed. Sensory evaluation was carried out for an appearance, smell, color, flavor and overall acceptability by semi- trained panelists using 9 points hedonic rating (1= dislike extremely, 9= like extremely).

In the Fig. 7, the percentage mentioned inside the bracket refers to the concentration of oleoresin.

The order of superiority can be summarized as;

General appearance: Sample C > Sample B > Sample A

Smell: Sample C > Sample B ≥ Sample A

Color: Sample C > Sample B > Sample A

Flavor: Sample C > Sample B > Sample A

Overall acceptance: Sample C > Sample B > Sample A

Sample C scored the highest in terms of all sensory attributes followed by sample B. Sample B and C were selected for further microbiological analysis.

Antimicrobial Activity Analysis

TVC and total ***Salmonella*** count were carried out to study the inhibition potency of the oleoresin extract. The reduction in the count of viable bacterial cells after incubation of significant duration with extracts from the study compound indicated the potential antibacterial activity against the pathogenic bacteria under the study.

Total Viable Count

Total viable counts (TVC) refers to the widely accepted measure of the general degree of microbial contamination and hygienic conditions of processing plants and slaughterhouses (Cohen et al., 2007) Minced meat with oleoresin concentrations of 0.07% and 0.1% and control were used for total plate count. Total plate count was determined on plate count agar and results were expressed in CFU/g.

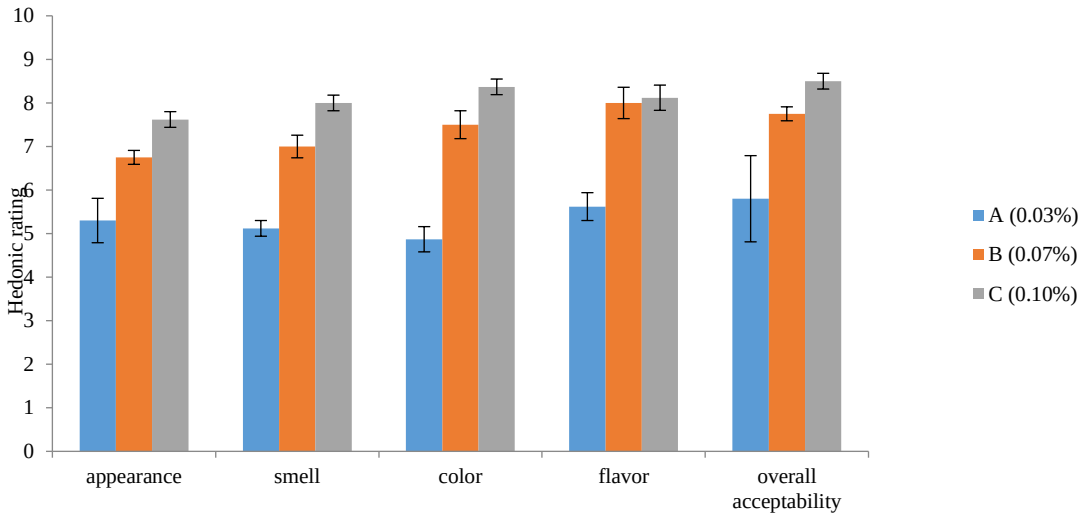

Fig. 7: Mean scores of the samples

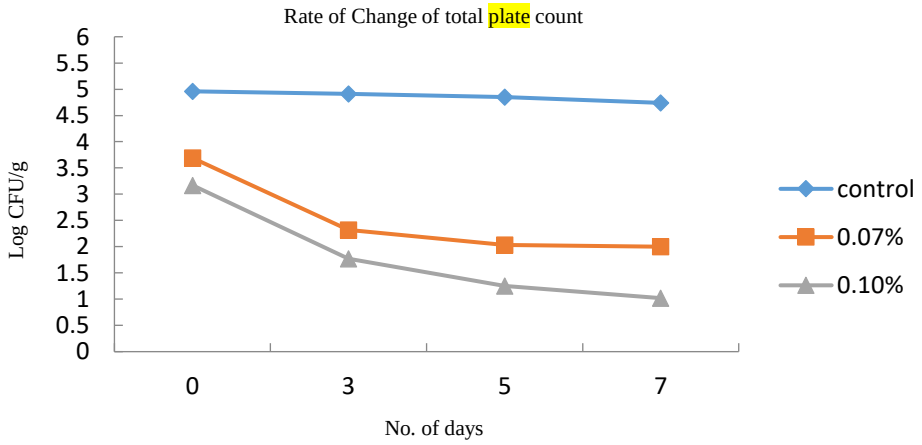

Fig. 8: Change in total plate count of raw minced chicken meat sample having different concentrations of oleoresin stored at 4ºC.

The change in total plate count of raw minced chicken meat stored at 4ºC within 7 days was calculated. Chicken samples infused with lower concentrations of oleoresin exhibited elevated logarithm values of Colony Forming Units (CFU) compared their counterparts with higher oleoresin concentrations as shown in Fig. 8. Despite this, both sets of samples demonstrated a noteworthy inhibition of the initial microbial load. Moreover, the sample containing a higher concentration of oleoresin showed a reduction in microbial load subsequently up to 7 days after the test.

The microbial load was found to be lower than the raw control sample and the microbiological standard (6.69 $\log_{10}$ cfu/g) on both the samples studied, indicating the efficacy of Jimbu extract in the prevention of microbial growth.

Analysis of Change in Salmonella Count

Salmonella count was determined by studying the growth of targeted bacteria on the xylose lysine deoxycholate agar and results were expressed in CFU/g.

The Fig. 9 illustrated how the Salmonella count changed during a 7-day period at 4ºC in three samples: those containing 0.07%, 0.1% oleoresin, and the control. The findings revealed complete inhibition by day 5 at 0.1% oleoresin concentration. Additionally, lower oleoresin concentrations exhibited full inhibition, slightly delayed, occurring by day 7. This indicated the effective antimicrobial properties of oleoresin on Salmonella growth, with higher concentrations indicating quicker and more potent inhibitory effects. The detailed analysis of *Salmonella* count is mentioned in Table 2 showing the significant reductions against *Salmonella growth* at both concentrations of oleoresins under study.

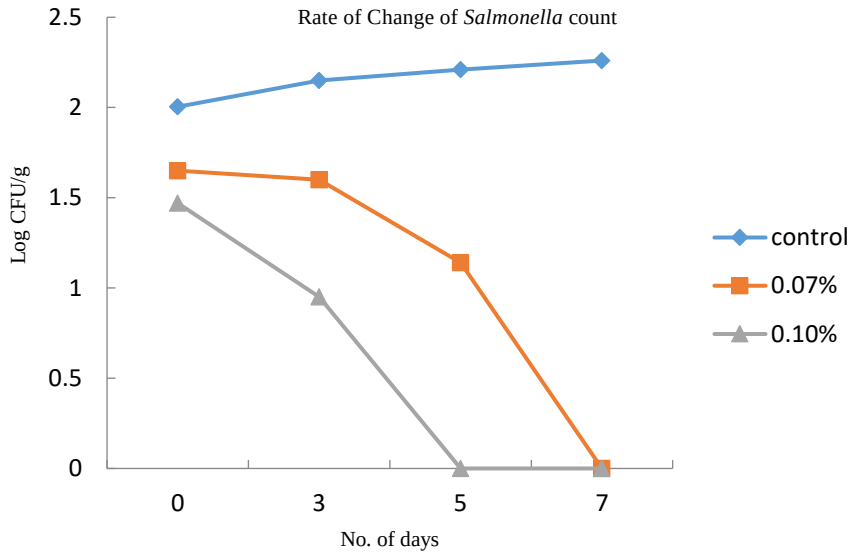

Fig. 9: Change in *Salmonella* count of raw minced chicken meat sample having different concentrations of oleoresin stored at 4ºC.

Table 2: Microbial load of minced chicken from initial day to day 7 for test of oleoresin.

		Days of incubation			
parameters		**0**	**3**	**5**	**7**
control	TPC(CFU/g)	4.96	4.92	4.85	4.74
	Salmonella (CFU/g)	2.0	2.16	2.21	2.27
0.07% oleoresin	TPC(CFU/g)	3.69	2.32	2.03	1.94
	Salmonella (CFU/g)	1. 70	1.60	1.14	No growth
0.10%oleoresin	TPC(CFU/g)	3.18	1.78	1.26	1.02
	Salmonella (CFU/g)	1.4789546	0.95424251	No growth	No growth

Conclusion

The use of medicinal plants in the treatment and prevention can be considered as prehistoric and their uses have been supported by the traditional optimization of their application in disease control. In this study, phytochemical analysis of *A. przewalskianum* (Jimbu*)* has been done along with its antimicrobial activity against *Salmonella* on minced chicken meat. The oleoresin obtained from Jimbu shows significant antimicrobial activity along with inhibition potency against *Salmonella*. This shows that the addition of oleoresin at a suitable concentration on minced chicken meat could effectively retard the growth of microbes and extend the shelf life. Similarly, the methanol extract showed increasing antioxidant activity with an increase in the methanol extract of Jimbu. Also, methanol extracts demonstrated the highest polyphenol, flavonoid and tannin content than petroleum ether extract. Therefore, further analysis of Jimbu for its antimicrobial potency for other pathogenic organisms could open the possibility of establishing this valuable medicinal plant as a raw source of medical formulations.

Author's Contribution

All authors have made equal contributions towards the present work, including manuscript preparation, revision of the manuscript for content and final approval of the manuscript.

Conflict of Interest

The authors declare that there is no conflict of interest with the present publication.

Acknowledgement

The authors are thankful to the Department of Food Technology, National College of Food Science and Technology and Central Department of Biotechnology, Tribhuvan University.

References

Abu Bakar MF, Mohamed M, Rahmat A, & Fry J (2009). Phytochemicals and antioxidant activity of different parts of bambangan (mangifera pajang) and Tarap (Artocarpus odoratissimus). *Food Chemistry* **113**(2): 479–483. DOI: 10.1016/j.foodchem.2008.07.081

Alam MN, Bristi NJ and Rafiquzzaman M (2013) Review on in vivo and in vitro methods evaluation of antioxidant activity. *Saudi Pharmaceutical Journal* **21**(2):143-152. DOI: 10.1016/j.jsps.2012.05.002

AOAC (2005) Evaluation of the TEMPO ® TVC for Enumeration of Total Aerobic Flora in a Variety of Foods, 69280.

Canadian Food Inspection Agency (CFIA) (2014) Repealed: Importation of Dried Plant Material (D-95-09, 2nd Revision). Ottawa, ON: Government of Canada.http://www.inspection.gc.ca/plants/plant-protection/directives/imports/d-95-09/eng

Cohen N, Ennaji H, Bouchrif B, Hassar M and Karib H (2007). Comparative study of microbiological quality of raw poultry meat at various seasons and for different slaughtering processes in Casablanca (Morocco). *J Appl Poul Res* **16**(4): 502-508. DOI: 10.3382/japr.2006-00061

Diniz do Nascimento L, Moraes AAB, Costa KSD, Pereira Galúcio JM, Taube PS, Costa CML, Neves Cruz J, de Aguiar Andrade EH and Faria LJG (2020) Bioactive Natural Compounds and Antioxidant Activity of Essential Oils from Spice Plants: New Findings and Potential Applications. *Biomolecules* **10**(7): 988. 10.3390/biom10070988

Galeotti F, Barile E, Curir P, Dolci M and Lanzotti V (2008) Flavonoids from carnation (*Dianthus caryophyllus*) and their antifungal activity; *Phytochemistry Letters* **1**(1): 44-48. DOI: 10.1016/j.phytol.2007.10.001

Huang X & Ahn DU (2019). Lipid oxidation and its implications to meat quality and human health. *Food science and biotechnology* **28**(5): 1275–1285. DOI: 10.1007/s10068-019-00631-7

IUCN Nepal (2000) National Register of Medicinal Plants. Katmandu: IUCN Nepal

Kapadiya DB, Dabhi BK and Aparnathi KD (2016). Spices and Herbs as a Source ofNatural Antioxidants for Food. *Int J Curr Microbiol. App Sci* **5**(7): 280–288. DOI: 10.20546/ijcmas.2016.507.029

Kaur G, Gupta V, Christopher AF and Bansal P (2016) Antioxidant potential of most commonly used vegetable-onion (Allium cepa L.). *Journal of Complementary and Alternative Medical Research* **1**(1):1-5.

Lin H-Y, Kuo Y-H, Lin Y-L and Chiang W (2009). Antioxidative Effect and Active Components from Leaves of Lotus (Nelumbo nucifera). *Journal of Agricultural and Food Chemistry*, ***57***(15), 6623–6629. DOI: 10.1021/jf900950z

Nepal RC. Status, Use and Management of Jimbu (Allium spp.): A Case Study from Upper Mustang, Nepal. 2006; May, ICIMOD, Himaldoc.

Peryam DR, Pilgrim FJ. Hedonic scale method of measuring food preferences. *Food Technology*. 1957; Vol 11, Suppl., 9-14.

Rajesh BR, Potty VP and Sreelekshmy SG (2016) Study of total phenol, flavonoids, tannin contents and phytochemical screening of various crude extracts of Terminalia catappa leaf, stem bark and fruit. *International Journal of Applied and Pure Science and Agriculture* **2**(6): 291-296.

Rezazi S, Abdelmalek S, Hanini S, Pina A, Ferrão P, Fournier J, et al. (2017) Kinetic Study and Optimization of Extraction Process Assessing the feasibility of using the heat demand-outdoor temperature function for a long-term district heat demand forecast. *Energy Procedia*. **139**: 98–104. DOI: 10.1016/j.egypro.2017.11.180

Rouger A, Tresse O and Zagorec M. Bacterial Contaminants of Poultry Meat: Sources, Species, and Dynamics. *Microorganisms* **5**(3): 50. 10.3390/microorganisms5030050

Samarth RM, Panwar M, Kumar M, Soni A, Kumar M and Kumar A (2008). Evaluation of antioxidant and radical-scavenging activities of certain radioprotective plant extracts. *Food Chemistry* **106**(2), 868–873. DOI: 10.1016/j.foodchem.2007.05.005

Samatha T, Shyamsundarachary R, Srinivas P and Ramaswamy N (2012). Quantification of total phenolic and total flavonoids content in extracts of Oroxylum indicum L. Kurz. *Asian J Pharmaceutical and Clinical Res* **5**(4): 177–179.

Shin SA, Joo BJ, Lee JS, Ryu G, Han M, Kim WY, Park HH, Lee JH and Lee CS (2020). Phytochemicals as Anti-Inflammatory Agents in Animal Models of Prevalent Inflammatory Diseases. *Molecules* (Basel, Switzerland), **25**(24): 5932. 10.3390/molecules25245932

Shrestha TB and Joshi RM (2000) National Register of Medicinal Plants. 20-26

Sultana B, Anwar F, Rafique Asi M and Ali Shahid Chatha S (2008). Antioxidant potential of extracts from different agro wastes: Stabilization of corn oil. *Grasas Y Aceites* **59**(3): 205–217. DOI: 10.3989/gya.2008.v59.i3.510

Taher Z and Sarmidi M (2015) Optimization Processing Parameters for Curcuma xanthorriza Oleoresin Yield and its Antioxidant Activity. *Int J Biotech* **4**(3): 97-102. DOI: 10.6000/1927-3037.2015.04.03.3

Unila S (2015) *OleoresinProcessing*. Retrievedfrom: http://staff.unila.ac.id/sugiharto/files/2015/03/Oleoresin-Processing.pdf

Walvekar Shubhada and Kaimal Prasanna (2014). Comparative study of effect of physical and chemical mutagens on phytochemical content and antioxidant activity of Aegle marmelos Corr. *International Journal of Advanced Research* **2**: 590-595.