REVIEW ARTICLE

Ananas comosus (L.) Merr.: A mini review of its therapeutic properties

Izzati Awang Rahman, Siti Nazrina Camalxaman, Azlin Sham Rambely, Norhisham Haron, Emida Mohamed*

Centre of Medical Laboratory Technology, Faculty of Health Sciences, Universiti Teknologi MARA, Cawangan Selangor Kampus Puncak Alam, 42300 Bandar Puncak Alam, Selangor, Malaysia

***Corresponding Author**

Emida Mohamed
Email: emida894@uitm.edu.my

Abstract:

Ananas comosus (L.) Merr. is a monocot perennial plant that belongs to the Bromeliaceae family. It is known as pineapple and is a common fruit in Malaysia. This review discusses its wide variety of phytochemical compounds and pharmacological properties. Based on the review, several parts of the *A. comosus* plant have been studied which include its stem, peel, crown, leaves and fruits. Alkaloids, α-amylase, acid and alkaline phosphatase, anthraquinones, bromelain, cardiac glycoside, coumarins, flavonoids, glycoside, inulin, napthoquinones, phenols, phytate, phytosterols, polyphenols, quinine, ribulose biphosphate carboxylase, saponin, steroids, sterols, tacorin, tannins, terpenoids and triterpenes have been identified from several parts of *A. comosus.* Due to the enormous number of bioactive compounds, various therapeutic properties and activities were recorded. Extracts of the plant has been proven to have medicinal effects such as protein anti-aggregation, wound healing, anti-proliferative, pro-apoptotic, anti-rheumatic, anti-inflammatory, antioxidant, antimicrobial, anti-diabetic, anti-coagulant, anthelminthic, anti-hyperglycemic, anti-plasmodial, anti-pyretic and cardioprotective properties.

Keywords: *Ananas comosus*, medicinal properties, phytochemical properties

1. INTRODUCTION

Herbs and herbal-derived drugs have for many decades played a key function in combating illness [1]. Today, many of modern chemical drugs still use medicinal plants as their main raw materials. Examples of such breakthrough are antimalarial and anticancer drugs [2]. Hao stated that around one-third of prescribed medicines came from botanical extracts and/or their variants [2]. Having said that, *Ananas comosus* or more commonly known as pineapple, is one of the plants that had been studied widely throughout the world to understand and explore its potential. This monocotyledon plant belongs to the Bromeliaceae family [3]. The pineapple has unique characteristics and due to its sweetness, only the fruits are taken for consumption. The other parts of the plant are often thrown away. According to Ketnawa et al. (2012), the waste from pineapple can be turned into value-added products [4].

Hence, the objective of the review is to compile available data on phytochemical compositionS and medicinal benefits of *A. comosus* based on previous studies. Studies on *A. comosus* have been conducted as early as 1936 until 2020 [5], [6]. However, the present review focuses only on research conducted for the past ten years. Available literature was focused on the fruit, peels and leaves of *A. comosus*. There were very limited studies on the core, crown, stem and bark of the plant.

2. BOTANY

The word pineapple is thought to originate from the pine cone due to their resemblance [3]. Thalip et al. (2015) stated that the name 'Ananas' was derived from its original native name in Brazil [7]. In Malay it is called nanas from the word 'nana'. The plant is a monocotyledon, which belongs to the genus of Ananas and family Bromeliaceae. There are three subfamilies in Bromoliaceae namely; Tillandsioideae, Bromeliodeae and Pitcarniodeae; in which *A. comosus* lies in the Bromeliodeae subfamily [8].

A. comosus is a tropical plant with herbaceous and monocot perennial plant characteristics. Having a unique structure and shape, its size varies from medium to large. Typically, the adult plant is approximately 1-2m high and 1-2m wide [9]. The major structures of pineapple morphology are its stem, leaves, peduncle, multiple fruit, crown, shoots, and roots (Figure 2.1). Several flowers are arranged helically along the axis, each develop a fleshy fruitlet which pushed against the fruitlets of neighbouring flowers, creating an appearance of a single fleshy fruit [9]. A fully mature pineapple plant has around 68-82 leaves, arranged in the form of a thick, compact rosette. The leaves are sword-like shaped and taper to the tip with length of about 5-20cm. The pineapple fruit is a compound fruit that develops through the fusion of many individual fruitlets [3]. The top of the fruit is known as the crown which function as an extension of the apical meristem and can be used as planting material [3]. Other parts of the plant that can be used for vegetative reproduction are the suckers, slips, butt and stumps.

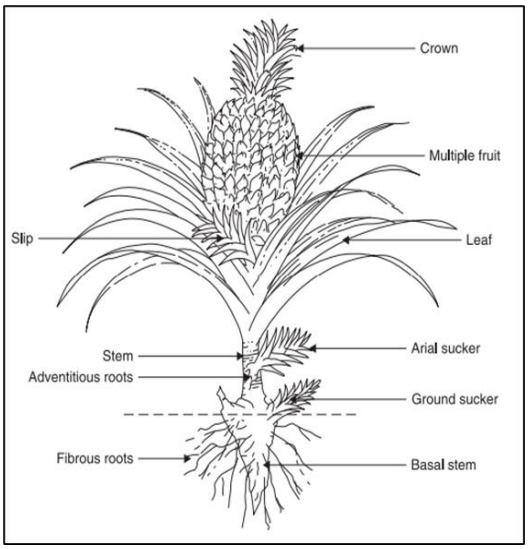

Figure 2.1 Morphological structure of *A. comosus*
Source: Hassan et al. (2011). Woodhead Publishing Limited

At present, there are about 30 pineapple cultivars worldwide that are cultivated under diverse environmental conditions in tropical regions. However, for commercial convenience, these cultivars are grouped into four groups. They are called "Red Spanish", "Pernambuco", "Queen" and "Smooth Cayenne" [10]. In Malaysia, there are several cultivars that can be found which are "Josapine", "Gandul", "Maspine", "N36", "Moris" and "MD-2".

3. PHYTOCHEMICAL PROPERTIES

Phytochemicals are non-nutritive plant chemicals with defensive and illness-preventive properties [11]. Usually, they are produced as chemicals and act as protection for the plants. From several studies on pineapple, it was found that a variety of phytochemical constituents are present in different parts of the pineapple as demonstrated in Table 2.1. However., even though quite similar studies were conducted, variation were seen in the results obtained. Gunwantrao et al. (2016) stated that these results are unlikely to match even if similar procedures have been used [12]. Presence or absence of certain phytochemical depends on the type of solvents used, nature of extraction process or climatic state of the region from where the pineapple were collected.

Previous studies on the stem of *A. comosus* conducted by Zaman et al. (2016) and Rahayu et al. (2017) have reported presence of bromelain and tacorin, respectively [13], [14]. In addition, saponin, flavonoids, phytosterol, carbohydrate, alkaloid, cardiac glycoside, amino acid and protein were also found in the stem [15].

Qualitative phytochemical screening of the peel extracts of *A. comosus* revealed the presence of protein, alkaloids, tannins, flavonoids, steroids, saponin, terpenoids, inulin, glycoside, phenolic compound, polyphenols, anthraquinones, coumarins, triterpenes, sterols, oxalate, phytate, quinine and carbohydrate [12], [16]-[20]. The total content of flavonoids and tannins were estimated as 1.76 ± 0.067mg/g and 4.52 ± 0.45mg/g, respectively [21]. Total flavonoid and phenolic content in the peel were studied using Aluminium Chloride Spectrophotometric and Folin Ciocalteau method [19]. Flavonoids was found higher in methanol extracts (860µg/ml) whereas phenolic was higher in ethanol extract (6930µg/mL). In addition, the presence of bromelain was also reported by other researcher [4].

Apart from the stem, the crown of *A. comosus* was also reported to contain bromelain [22]. The crown extract was prepared using purified water as solvent. Bromelain was then purified using Preparative HPLC (Agilent) using cation exchange resin column. Protein profiling of the crown showed the presence of ribulose biphosphate carboxylase alongside with other enzymes such as peroxidase, acid and alkaline phosphatase, nuclease and α-amylase [23].

When the leaves were analysed, presence of flavonoids, terpenoids, phytosterol, amino acids, phenols, tannins, carbohydrates, glycosides, proteins, alkaloids and saponins were reported [15], [24]-[26]. In addition, Mondal et al. (2011) reported on the presence of triterpenoids, steroids, alkaloids, saponins, glycosides and carbohydrates in the extracts of petroleum ether, chloroform and methanol [27]. Work conducted by Jovanović et al. (2018) found the highest amount of phenols found in the pineapple leaves extract were that prepared by using 60% methanol (25.94 ± 3.54 mg GAE/g) in comparison to 80% methanol (22.99 ± 2.36 mg GAE/g) [28].

Fruits of *A. comosus* were also analysed for its phytochemical constituents. When extracted with seven different solvents namely ethanol, methanol, ethyl acetate, acetone, n-Hexane, chloroform and distilled water, the phytochemical compounds yielded were saponins, tannins, steroids, flavonoids, terpenoids, napthoquinone, inulin, alkaloids, phenols and amino acids [29]. Similarly, presence of terpenoids, flavonoid, phytosterol, alkaloid, cardiac glycoside, amino acid, protein, polyphenols, saponins, and bromelain were reported by other researchers [15], [30]. Quantitative analysis revealed the phenolic and flavonoid activity were found higher in methanol extract with 51.1 ± 0.2mg/g and 55.2 ± 0.2mg/g, respectively in comparison to water and ethyl extracts [31]. Table 2.1 summarizes the phytochemicals present in the different parts of *A. comosus.*

4. MEDICINAL PROPERTIES

Study by Zaman et al. (2016) tried to investigate the possibility of using bromelain extracted from the stem of *A. comosus* to prevent protein aggregation [13]. Protein aggregation can be seen in neurodegenerative disorder such as in prion disease and Alzheimer's disease that is caused by several forms of amyloidosis. Studying the effects of stem bromelain on pH may be useful for the development of new drug. Their group tried to look at the effect of bromelain on fibril formation at various pH and it was found that at pH 2, no aggregation was formed. In contrast, at pH 10, where negative charge was produced from bromelain, aggregation of proteins was detected. In 2017, Rahayu et al. did an *in vivo* study to look at the effect of tacorin in the stem extract of *A. comosus* on wound healing [14]. They managed to demonstrate that the presence of tacorin produced faster rate of recovery in wounds of rats.

Table 2.1: Summary of phytochemical components reported present in *A. comosus*.

Phytochemical properties	Stem	Peel	Crown	Leaves	Fruits
α-amylase	-	-	+	-	-
Acid phosphatase	-	-	+	-	-
Alkaline phosphatase	-	-	+	-	-
Alkaloid	+	+	-	+	+
Amino acids	+	-	-	+	+
Anthraquinones	-	+	-	-	-
Bromelain	+	+	+	-	+
Carbohydrate	+	+	-	+	-
Cardiac glycoside	+	-	-	-	+
Coumarins	-	+	-	-	-
Flavonoids	+	+	-	+	+
Glycoside	-	+	-	+	-
Inulin	-	+	-	-	+
Napthoquinones	-	-	-	-	+
Nuclease	-	-	+	-	-
Oxalate	-	+	-	-	-
Peroxidase	-	-	+	-	-
Phenols	-	+	-	+	+
Phytate	-	+	-	-	-
Phytosterols	+	-	-	+	+
Polyphenols	-	+	-	-	+
Protein	+	+	-	+	+
Quinine	-	+	-	-	-
Ribulose biphosphate carboxylase	-	-	+	-	-
Saponin	+	+	-	+	+
Steroids	-	+	-	+	+
Sterols	-	+	-	-	-
Tacorin	+	-	-	-	-
Tannins	-	+	-	+	+
Terpenoids	-	+	-	+	+
Triterpenes	-	+	-	+	-

+: detected; -: not detected

The amino acid composition of tacorin, when analyzed using HPLC, showed presence of glycine, proline, glutamine and alanine at 0.91%, 0.89%, 0.71% and 0.65%, respectively. The amino acid mentioned above were important in wound healing process. While an *in vitro* study conducted on mammalian cells showed an increase in the TNF-β expression which is important in tissue remodelling and the TNF-α indicated that the excessive inflammation was suppressed by tacorin. P13K/Akt and MAP kinase pathways are two significant pathways for the regulation of tumor cell growth and apoptosis. Therefore, in order to examine the potential anti-proliferative and pro-apoptotic activities of bromelain, these two pathways were studied [32]. Their findings showed that bromelain downregulated phosphorylation of Akt, ERK_1, and ERK_2 in Caco-2 cells indicating potential role of bromelain in these pathways. Bromelain appeared to be able to promote apoptotic cell death in tumor cells and decrease tumor cell proliferation by suppressing phosphorylation of Akt and $ERK_{1/2}$, respectively.

In 2016, Kargutkar and Brijesh studied on the methanolic extract of *A. comosus* peel (MEFP) using Freund's induced arthritis model and found that the extract possessed anti-rheumatic, anti-inflammatory and antioxidant properties [21] as shown in Table 2.2. The extract demonstrated a substantial decrease in inflammation measured at 100mg/kg b.w. and 500mg/kg b.w., which was equivalent to the effect seen by normal medication using prednisolone at a concentration of 10mg/kg b.w. It was also found that when MEFP was administered in the rats, there were significant reduction of CRP and PGE_2 level; which led to the suggestion on the potential of anti-inflammatory properties

Table 2.2: Summary of medicinal properties of *A. comosus*

Medicinal properties	Stem	Peel	Crown	Leaves	Fruits
Anti-coagulant	-	+	-	-	-
Anti-diabetic	-	+	-	+	-
Anthelminthic	-	+	-	+	-
Anti-hyperglycemic	-	-	-	+	-
Anti-inflammatory	-	+	-	+	-
Antimicrobial	-	+	+	+	+
Antioxidant	-	+	+	+	+
Anti-plasmodial	-	-	-	-	+
Anti-proliferative	+	-	-	-	-
Anti-pyretic	-	-	-	-	+
Anti-rheumatic	-	+	-	-	-
Wound healing	+	-	-	-	-
Pro-apoptotic	+	-	-	-	-

+: detected; -: not detected

in *A. comosus*. The increase of the SOD, CAT and GPx production in the liver, kidney and spleen when treated with MEFP demonstrates its protective role against oxygen-derived free radicals by restoring the function of radical scavenging enzymes. The findings were close to those seen when using prednisolone.

The peels were also shown to possess antioxidant activity with reducing power that reduced Fe^{3+} to Fe^{2+} [20]. In 2018, Jovanović et al. (2018) revealed that the highest antioxidant capacity was found in the methanol peel extract [28]. Beside methanol extract, aqueous and ethanol extracts also demonstrated antioxidant activity when measured by DPPH radical scavenging assay, hydrogen peroxide scavenging activity, and total antioxidant activity [16]. Das et al. (2019) also reported that *A. comosus*-silver nanoparticles (AC-AgNPs) exhibited possible antioxidant activity [33].

Apart from anti-inflammatory and antioxidant properties, the peel ethanolic extract also demonstrated maximum zone inhibition against pathogenic bacterial strains such as *Klebsiella pneumonia*, *Pseudomonas aeruginosa*, *Bacillus subtillis* and *Xanthomonas axonopodis pv. Malvacearum* [12]. Growth of *Escherichia coli* and *Bacillus amyloliquifaciens* were found to be inhibited by ethanol, methanol and n-hexane peel extracts while *Staphylococcus aureus* and *Pseudomonas aeruginosa* were inhibited by extracts prepared by ethanol and methanol [19]. In 2017, Sharma et al. reported that the peels showed antimicrobial reaction when tested with *Escherichia coli* and *Staphylococcus aureus* [20]. The aqueous and ethanolic extracts of *A. comosus* peel also demonstrated remarkable inhibition of *Bacillus cereus*, *Escherichia coli*, *Klebsiella pneumonia* and methanol and *Staphylococcus aureus* [17]. Besides, mild antibacterial activity against *Listeria monocytogenes* which were examined in food were also reported by previous researchers [33].

In 2019, Das et al. reported that *A. comosus*-silver nanoparticles (AC-AgNPs) has a strong anti-diabetic ability which was highly efficient even at a very low concentration [33]. It also demonstrated strong dose-dependent cytotoxic activity against the HepG2 cancer cells. The ethanolic extracts of *A. comosus* peel was also found to have an anticoagulant property when tested against ICR mice [34]. The pineapple peel extract demonstrated longer average coagulation time and the *p*-value showed significance differences compared to other plants that were tested. In addition, the aqueous extract was successfully used as an anthelmintic agent against *Haemonchus contortus in vitro*

but demonstrated a reduced anthelmintic efficacy when tested *in vivo* [35].

The crown of *A. comosus* was found to contain antioxidant capacity that was measured by using DPPH assay [28]. Earlier, study by Dutta and Bhattacharyya (2013) tried to investigate the potential of antimicrobial and toxicity of the crown extracts [23]. *Saccharomyces cerevisiae*, *Staphylococcus aureus*, *Bacillus subtilis*, *Candida albicans* and *Escherichia coli* showed significant microbial growth inhibition, thus proving that crown extracts possessed bacteriostatic and fungistatic components. As for its toxicity effects that were evaluated using both acute and sub-acute toxicity studies, there were no sign of mortality and toxicity against the adult female rats. It was found that no significant change in serum biomarker concentrations of ALT, AST and ALP were shown when compare to the control groups.

In 2010, Kataki found the presence of antibacterial, antioxidant and anthelminthic activity in ethanolic extracts of *A. comosus* leaves [24]. The minimum inhibitory concentration (MIC) values of *Staphylococcus aureus*, *Salmonella typhi* and *Escherichia coli* were 0.3125mg/ml, 0.625mg/ml and 1.25mg/ml, respectively. As for its antioxidant property, at concentration of 250µg/ml, stronger antioxidant activity was seen in *A. comosus* extract (57.2%) compared to α-tocopherol (26%) but lower than that of butylated hydroxyanisole (96.8%). In the antihelmithic assay, the extract was tested against *Pheretima posthuman*. It was found that at 100mg/ml concentration, the extract took the least time to cause paralysis and worm death when compared to piperazine citrate and albendazole. Similarly, Ahmed et al. (2013) also reported anthelminthic activity in *A. comosus* leaves extracts [36]. In their work, ethanol, dichloromethane and aqueous extracts of *A. comosus* were proven to inhibit larval development of *H. contortus in vitro*. Okokon et al. (2019) also proved that the ethanol extract possessed anti-plasmodial and anti-pyretic properties when tested against early *Plasmodium berghei* infection in mice and D-amphetamine-induced-pyrexia, respectively [37].

In 2011, Mondal et al. stated that the methanol leaf extracts possessed acute anti-inflammatory property against rats due to the presence of its bioactive compounds [27]. This activity was also supported by a study by other researcher [26]. *A. comosus* leaf extracts (ALE) exhibited potential anti-inflammatory activities as TNF-α, IL-1β, PGE_2 and ROS decrease significantly. Furthermore, the ALE extracts also showed reduce in paw size in the Carrageenan-induced edema experiment. Besides, *A. comosus* is also beneficial as anti-hyperglycemic and anti-diabetic agents when tested against induced diabetic rats [25], [38]. The hydro-alcoholic extract of *A. comosus* proved to be comparable to the standard drug glibenclamide as both substantially reduced the fasting blood glucose and elevated the body weight in streptozotocin induced diabetic rats [38]. Reduce in blood glucose level as much as 46.9% was seen at the highest dose of methanol extract of *A. comosus* leaves (400mg) which was comparable to standard drug, glibenclimade [25].

Study by Zharfan et al. (2017) proved that the fruit of *A. comosus* contain antimicrobial properties [30]. The presence of bromelain and saponin in the extract resulted in its ability to destruct *P. aeruginosa* cell wall and membranes. According to Eshamah et al. (2013), bromelain is speculated to trigger protein breakdown in the bacterial membrane, causing damage and cell death [39]. It was suggested that bromelain behaves via an associated mechanism to destabilize the outer membrane of gram-negative protein-containing bacteria. Bromelain is assumed to deteriorate protein in the surface membrane, which subsequently weakens the cell wall, resulting in cell spillage, swells the cell, and harms the cell. Saponins, on the other hand, enhances the permeability of the bacterial cell membrane, inducing modification of the shape and function of the membrane, weakening the surface tension of the cell wall, and enabling antibacterial compounds to simply reach the cells and interact with cell metabolism whilst denaturing proteins on the membrane [40]. Recent research by Kaushik and Kundu (2018) also found that the fruit possess antimicrobial activity [29]. Aqueous extracts were found to inhibit the growth of *Escherichia coli*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* with inhibition length of 13mm, 10mm and 13mm, respectively.

In addition, *A. comosus* fruit extract was also proven to have antioxidant ability [29]. Methanolic extract showed the highest antioxidant activity compared to ethyl acetate and water extracts [31]. In 2014, Saxena and Panjwani demonstrated that the level of cholesterol, triglycerides, LDL and VLDL in isoproterenol induced myocardial infarcted rats were substantially decrease in the serum, while HDL levels were increased when fed with hydro-alcoholic extract of *A. comosus* [41]. In addition, since *A. comosus* has high antioxidant and bromelain capacity, which can preserve the integrity of the cell membrane, it managed to boost the cardiac systolic/diastolic instability of isoproterenol. The results showed that the extract was capable in minimizing the severity of myocardial injury and greatly counteracting the oxidative stress in rats during myocardial infarct caused by isoproterenol.

5. CONCLUSION

A .comosus or commonly known as pineapple is one of the major plants found in Malaysia. A lot of studies have been conducted to explore beneficial attributes from various parts of the *A. comosus* plant. The stem, peel, crown, leaves and fruits have been used to prepare extracts and tested for various therapeutic activities. MAJORITY of the researchES were performed by *in vivo* method. As such, exploration of the reported properties using cell culture or animal model is necessary to ensure its safety for human consumption and usage. In conclusion, therapeutics properties of *A. comosus* had been scientifically proven which offer tremendous potentials to be developed into various pharmaceutical products.

REFERENCES

[1] T. C. Okoye, P. F. Uzor, C. A. Onyeto, and E. K. Okereke, "Safe African Medicinal Plants for Clinical Studies," in *Toxicological Survey of African Medicinal Plants*, Elsevier Inc., 2014, pp. 535–555.

[2] D.-C. Hao, *Genomics and Evolution of Medicinal Plants*. Elsevier Inc., 2019.

[3] S. W. Wijeratnam, "Pineapple," *Encycl. Food Heal.*, pp.

380–384, 2015, doi: 10.1016/B978-0-12-384947-2.00547-X.

[4] S. Ketnawa, P. Chaiwut, and S. Rawdkuen, "Pineapple wastes: A potential source for bromelain extraction," *Food Bioprod. Process.*, vol. 90, no. 3, pp. 385–391, 2012, doi: 10.1016/j.fbp.2011.12.006.

[5] K. R. Kerns, J. L. Collins, & H. Kim, "Developmental Studies of the Pineapple *Ananas comosus* (L) Merr. L. Orgin and Growth of Leaves and Inflorescence," *The New Phytologist*, vol. 35, no. 4, pp. 305-317, 1936.

[6] A. Larrea-Sarmiento *et al.*, "Identification and complete genomic sequence of a novel sadwavirus discovered in pineapple (*Ananas comosus*)," *Arch. Virol.*, vol. 165, no. 5, pp. 1245–1248, 2020, doi: 10.1007/s00705-020-04592-9.

[7] A. A. Thalip, P. S. Tong, and C. Ng, "The MD2 'Super Sweet' pineapple (*Ananas comosus*)," *Utar Agric. Sci. J.*, vol. 1, no. 4, pp. 14–17, 2015, [Online]. Available: http://eprints.utar.edu.my/1982/1/The_MD2_(Super_Sweet)_pineapple_(Ananas_comosus).pdf.

[8] Office of the Gene Regulator Technology, *The Biology of Ananas comosus var . comosus (Pineapple)*, vol. 2, no. February. 2008.

[9] A. Hassan, Z. Othman, and J. Siriphanich, *Pineapple (Ananas comosus L. Merr.)*, vol. 4. Woodhead Publishing Limited, 2011.

[10] N. Wali, *Pineapple (Ananas comosus)*, vol. 1. Elsevier Inc., 2018.

[11] Phytochemicals.info. n.d. *Phytochemicals*. [online] Available at: <https://www.phytochemicals.info/> [Accessed 16 July 2020].

[12] B. B. Gunwantrao, S. K. Bhausaheb, B. S. Ramrao, and K. S. Subhash, "Antimicrobial activity and phytochemical analysis of orange (*Citrus aurantium* L.) and pineapple (*Ananas comosus* (L.) Merr.) peel extract," *Ann. Phytomedicine An Int. J.*, vol. 5, no. 2, pp. 156–160, 2016, doi: 10.21276/ap.2016.5.2.22

[13] M. Zaman, A. Ehtram, S. K. Chaturvedi, S. Nusrat, and R. H. Khan, "Amyloidogenic behavior of different intermediate state of stem bromelain: A biophysical insight," *Int. J. Biol. Macromol.*, vol. 91, pp. 477–485, 2016, doi: 10.1016/j.ijbiomac.2016.05.107.

[14] P. Rahayu, L. Agustina, and R. R. Tjandrawinata, "Tacorin, an extract from *Ananas comosus* stem, stimulates wound healing by modulating the expression of tumor necrosis factor α, transforming growth factor β and matrix metalloproteinase 2," *FEBS Open Bio*, vol. 7, no. 7. pp. 1017–1025, 2017, doi: 10.1002/2211-5463.12241.

[15] P. A. Mary Helen, D. S. Teena, J. G. Godwin Jacob, J. Jinto James, and C. Anitha, "Preliminary Phytochemical Screening and Antioxidant Activity of Leaf, Stem and Fruit *Ananas comosus*," *World J. Pharm. Res.*, vol. 8, no. 5, pp. 1407–1416, 2019, doi: 10.20959/wjpr20195-14722.

[16] J. X. Agnes and A. Anusuya, "Phytochemical screening and in vitro antioxidant activity of *Ananas comosus*," *Int. J. Res. Pharmacol. Pharmacother.*, vol. 5, no. 2, pp. 162–169, 2016.

[17] A. P. Dabesor, A. M. Asowata-Ayodele, and P. Umoiette, "Phytochemical Compositions and Antimicrobial Activities of *Ananas comosus* Peel (M.) and *Cocos nucifera* Kernel (L.) on Selected Food Borne Pathogens," *Am. J. Plant Biol.*, vol. 2, no. 2, pp. 73–76, 2017, doi: 10.11648/j.ajpb.20170202.15.

[18] J. K. Dzotam and V. Kuete, "Antibacterial Activity Of Methanol Extracts From Six Cameroonian Edible Plants *Camellia sinensis*, *Mangifera indica*, *Moringa oleifera*, A*nanas comosus* , *Triumphetta pentandra* and *Artocarpus heterophyllus* Against MDR Gram-Negative Phenotypes," *African J. Integr. Heal.*, vol. 7, no. 2, pp. 49–56, 2017.

[19] Y. S. Namrata and T. Sharma, "Anti-Microbial, Anti-Oxidant Activity and Phytochemical Screening of Polyphenolic Flavonoids Isolated from Peels of *Ananas comosus*," *Int. J. Eng. Res. Technol.*, vol. 6, no. 9, pp. 285–298, 2017, doi: 10.17577/ijertv6is090176.

[20] G. Sharma, I. Vivek, A. K. Gupta, D. Ganjewala, C. Gupta, and D. Prakash, "Phytochemical composition, antioxidant and antibacterial potential of underutilized parts of some fruits," *Int. Food Res. J.*, vol. 24, no. 3, pp. 1167–1173, 2017.

[21] S. Kargutkar and S. Brijesh, "Anti-rheumatic activity of *Ananas comosus* fruit peel extract in a complete Freund's adjuvant rat model," *Pharm. Biol.*, vol. 54, no. 11, pp. 2616–2622, 2016, doi: 10.3109/13880209.2016.1173066.

[22] K. Z. Nadzirah, S. Zainal, A. Noriham, I. Normah, and A. M. S. Roha, "Physico-Chemical Properties of Pineapple Crown Extract Variety N36 and Bromelain Activity in Different Forms," *APCBEE Procedia*, vol. 4, pp. 130–134, 2012, doi: 10.1016/j.apcbee.2012.11.022.

[23] S. Dutta and D. Bhattacharyya, "Enzymatic, antimicrobial and toxicity studies of the aqueous extract of *Ananas comosus* (pineapple) crown leaf," *J. Ethnopharmacol.*, vol. 150, no. 2, pp. 451–457, 2013, doi: 10.1016/j.jep.2013.08.024.

[24] M. S. Kataki, "Antibacterial activity, *in vitro* antioxidant activity and anthelmintic activity of ethanolic extract of *Ananas comosus* L. tender leaves," *Pharmacologyonline*, vol. 2, pp. 308–319, 2010.

[25] M. M. Faisal, F. M. M. Hossa, R. Shahnaz, A. B. M. Anwarul Bashar, H. Shahadat, and R. Mohammed, "Effect of methanolic extract of *Ananas comosus* leaves on glucose tolerance and acetic acid-induced pain in Swiss albino mice," *World J. Pharm. Res.*, vol. 3, no. 8, pp. 24–34, 2014, [Online]. Available: www.wjpr.net.

[26] S. Kargutkar and S. Brijesh, "Anti-inflammatory evaluation and characterization of leaf extract of *Ananas comosus*," *Inflammopharmacology*, vol. 26, no. 2, pp. 469–477, 2018, doi: 10.1007/s10787-017-0379-3.

[27] S. Mondal, S. Bhattacharya, J. N. Pandey, and M. Biswas, "Evaluation of acute anti-inflammatory effect of *Ananas comosus* leaf extracts in rats," *Pharmacologyonline*, vol. 3, pp. 1312–1315, 2011.

[28] M. Jovanović *et al.*, "Antioxidant capacity of pineapple (*Ananas comosus* (L.) Merr.) extracts and juice," *Lek. sirovine*, vol. 38, pp. 27–30, 2018, doi: 10.5937/leksir1838027j.

[29] J. Kaushik and N. Kundu, "Phytochemical Screening , Anti-oxidant and Anti-Microbial Activity of Polyphenolic Flavonoids Isolated from fruit of *Ananas comosus* in various solvents," *Int. J. Sci. Resesarch Publ.*, vol. 8, no. 2, pp. 31–55, 2018.

[30] R. S. Zharfan, P. B. Purwono, and A. Mustika, "Antimicrobial Activity of Pineapple (*Ananas Comosus* L. Merr) Extract Against Multidrug-Resistant of *Pseudomonas aeruginosa*: An *in vitro* Study," *Indones. J. Trop. Infect. Dis.*, vol. 6, no. 5, pp. 118–123, 2017, doi: 10.20473/ijtid.v6i5.4159.

[31] M. A. Hossain and S. M. M. Rahman, "Total phenolics, flavonoids and antioxidant activity of tropical fruit pineapple," *Food Res. Int.*, vol. 44, no. 3, pp. 672–676, 2011, doi: 10.1016/j.foodres.2010.11.036.

[32] B. Romano *et al.*, "The chemopreventive action of bromelain, from pineapple stem (*Ananas comosus* L.), on colon carcinogenesis is related to antiproliferative and proapoptotic effects," *Mol. Nutr. Food Res.*, vol. 58, no. 3, pp. 457–465, 2014, doi: 10.1002/mnfr.201300345.

[33] G. Das, J. K. Patra, T. Debnath, A. Ansari, and H. S. Shin, "Investigation of antioxidant, antibacterial, antidiabetic, and cytotoxicity potential of silver nanoparticles synthesized using the outer peel extract of *Ananas comosus* (L.)," *PLoS ONE*, vol. 14, no. 8. 2019, doi: 10.1371/journal.pone.0220950.

[34] A. R. Arenajo *et al.*, "*In Vitro* Anticoagulant Potential of *Caulerpa* sp . ('Lato'), *Eucheuma* sp . ('Guso'), *Ananas comosus* ('Pineapple') Peeling and *Psidium guajava* ('Guava') Leaf Extracts in ICR Mice," *J. Agric. Technol. Manag.*, vol. 21, no. 1, pp. 30–34, 2018.

[35] L. F. Domingues *et al.*, "*In vitro* and *in vivo* evaluation of the activity of pineapple (*Ananas comosus*) on *Haemonchus contortus* in Santa Inês sheep," *Vet. Parasitol.*, vol. 197, pp. 263–270, 2013, doi: 10.1016/j.vetpar.2013.04.031.

[36] M. Ahmed, M. D. Laing, and I. V. Nsahlai, "*In vitro* anthelmintic activity of crude extracts of selected medicinal plants against *Haemnchus contortus* from sheep," *J. Helminthol.*, vol. 87, no. 2, pp. 174–179, 2013, doi: 10.1017/S0022149X1200020X.

[37] J. E. Okokon, K. N. Opara, J. A. Udobang, and H. K. Bankehde, "*In vivo* antiplasmodial and antipyretic activities of ethanol leaf extract of *Ananas comosus* (L.) Merr," *Trop. J. Nat. Prod. Res.*, vol. 3, no. 7, pp. 240–245, 2019, doi: 10.26538/tjnpr/v3i7.5.

[38] A. B. Vuyyuru, M. Govindrao, R. C. S. Reddy, B. Harish, J. Vishwanath, and G. Amarnath Reddy, "Antidiabetic activity of hydroalcoholic extract of *Ananas comosus* L. Leaves in streptozotocin induced diabetic rats.," *Int. J. Pharm.*, vol. 2, no. 1, pp. 142–147, 2012.

[39] H. Eshamah, I. Han, H. Naas, J. Rieck, and P. Dawson, "Bactericidal Effects of Natural Tenderizing Enzymes on *Escherichia coli* and *Listeria monocytogenes*," *Journal of Food Research*, vol. 2, no. 1, 2013, doi: 10.5539/jfr.v2n1p8

[40] C. Y. Karlina, I. Muslimin, and G. Trimulyono, "Aktivitas Antibakteri Ekstrak Herba Krokot (*Portulaca oleraceae* L.) terhadap *Staphylococcus aureus* dan *Escherichia coli*," *Lentera Bio*, vol. 2, no. 1, pp. 87-93, 2013

[41] P. Saxena and D. Panjwani, "Cardioprotective potential of hydro-alcoholic fruit extract of *Ananas comosus* against isoproterenol induced myocardial infraction in Wistar Albino rats.," *J. Acute Dis.*, vol. 3, no. 3, pp. 228–234, 2014, doi: 10.1016/s2221-6189(14)60051-2.