A review study on the anti-*trichomonas* activities of medicinal plants

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ABSTRACT

The parasitic diseases represent the most important health risk, especially in underdeveloped countries where they have a deep impact on public health. Trichomoniasis is a prevalent non-viral sexually transmitted disease, and a significant amount of new cases are identified each year globally. Furthermore, the infection is linked with serious concerns such as pregnancy outcomes, infertility, predisposition to cervical and prostate cancer, and increased transmission and acquisition of HIV. The therapy is restricted, adverse effects are often observed, and resistance to the drugs is emerging. Based on this, a new treatment for trichomoniasis is necessary. Natural products represent a rich source of bioactive compounds, and even today, they are used in the search for new drugs. Additionally, natural products provide a wide variety of leadership structures that can be used by the pharmaceutical industry as a template in the development of new drugs that are more effective and have fewer or no undesirable side effects compared to current treatments.

This review focuses on the medicinal plants that possess anti-trichomonal activity *in vitro* or *in vivo*. An electronic database search was carried out covering the last three decades, i.e., 1990–2020. The literature search revealed that almost a dozen isolated phytoconstituents are being explored globally for their anti-trichomonal activity. Simultaneously, many countries have their own traditional or folk medicine for trichomoniasis that utilizes their native plants, as a whole, or even extracts. This review focuses mainly on the human parasite *Trichomonas vaginalis*. However, at some points mention is also made to *Tritrichomonas foetus* that causes trichomoniasis in animals of high veterinary and economical interest. We will focus on the plants and plant-based compounds and their anti-trichomonal activity. The literature search highlighted that there are abundant compounds that possess anti-trichomonal activity; however, in-depth *in-vivo* evaluation of compounds and their clinical evaluation has not been undertaken. There is a critical need for new anti-trichomonal compounds, and focused research on phytoconstituents can provide the way forward.

1. Introduction

1.1. Trichomonads and trichomoniasis pathogenesis

Trichomonads are anaerobic flagellated protists infecting many vertebrate and invertebrate species, fit into the large and diverse groups of Trichomonadea and Tritrichomonadea (Fig. 1).

Five species of trichomonads infect humans. They are *Trichomonas vaginalis,* found in the urogenital tract; *Trichomonas tenax,* localized in the oral cavity; *Pentatrichomonas hominis; Tetratrichomonas* sp, in the respiratory tract of humans (Dong et al., 2019) and digestive tract of several animals (Cepicka et al., 2006). and *Dientamoeba fragilis*, both found in the digestive tract (Maritz et al., 2014). *P. hominis* and *D. fragilis* cause gastrointestinal symptoms like abdominal pain and diarrhea, and infect domestic animals like cats and farm mammals, demonstrating non-human selectivity and zoonotic origin. Additionally, trichomonad species causing infections in birds and animals, and thus having veterinary importance, are *Trichomonas gallinae, Tetratrichomonas gallinarum,*

and *Histomonas meleagridis*, and *Tritrichomonas foetus* (Amin et al., 2014).

The only species with well-established pathogenic potential in humans is *T. vaginalis*. Trichomoniasis is the most common non-viral sexually transmitted urogenital infection that affects both men and women. Caused by flagellated protozoan parasite *Trichomonas vaginalis*, it mainly affects women's vagina, showing clinical symptoms like vaginal discharge, often yellow or green, vulvovaginal irritation, and dysuria. Men with trichomoniasis may not show any clinical signs; however, the urethra is the most common infection site, causing urethral irritation, discharge, or mild burning after urination or ejaculation and swelling of the prostate gland (Rein, 2020). Humans are the only host for the *T. vaginalis* parasite existing only in trophozoite form that uses adhesion proteins to bind to squamous epithelial cells. Soluble and membrane-associated enzymes having phospholipase A activity cause lysis of nucleated cells, resulting in microulcerations and microscopic hemorrhages of the vaginal wall and exocervix. Although humoral and cell-mediated immune-inflammatory responses are generated, repeated, and persistent infection occurs, indicating non-protective immunity (Meites and Workowski, 2018).

Pulmonary trichomoniasis, an opportunistic infection caused by *T. tenax,* is generally harmless; however, it may become serious in patients with poor oral hygiene. Periodontal diseases like gingivitis and periodontitis, resulting from disruption of the host–oral microbiome homeostasis, often trigger *T. tenax* infection. The flagellate can adhere to epithelial cells and produce diverse enzymes that cause periodontal breakdown (Bisson et al., 2019). It is generally observed as a mixed infection with chronic purulent or necrotic lung diseases, such as lung abscess or bronchiectasis (Hersh, 1985).

Treatment of trichomoniasis, for the last several decades, involves 5′-nitroimidazoles, e.g., metronidazole, tinidazole, ornidazole, and nimorazole. Metronidazole low-dose vaginal preparations are not effective for trichomoniasis; hence high-dose vaginal suppositories are used. The male partner(s) should also be treated simultaneously, even if asymptomatic. Metronidazole or tinidazole 2 g orally as a single dose of metronidazole 500 mg orally twice daily for seven days is the treatment option. Based on the observations of a multicentre, open-label, randomized controlled trial in women positive for *T. vaginalis* infection, 7-days low dose metronidazole is recommended as a primary treatment (Kissinger et al., 2018). Single-dose is less effective and is associated with side effects than the 1-week treatment, and it is preferred because

of patient compliance and lower cost of therapy (Seña et al., 2014). *T. tenax* is morphologically close to *T. vaginalis* and is sensitive to anti-parasitic treatment with tinidazole (Kurnatowska and Kurnatowski, 1999) as *T. vaginalis* (Butt and Tirmizi, 2018).

1.2. Challenges to the effective management of trichomoniasis

Although curable, trichomoniasis and its treatment are often challenging because of the drug's side effects. Generally, adverse effects include nausea, vomiting, constipation, cramping, and metallic taste. Other adverse effects include peripheral neuropathy, seizures, fatigue, dizziness, headache, and leukopenia (Wendel and Workowski, 2007). Additionally, trichomonas is increasingly associated with other health complications like pelvic inflammatory disease and cervical cancer. Preterm births, low birth weights, stillbirth, neonatal death, sexual transmission, and acquisition of HIV infection are strongly associated with trichomoniasis (Hirt et al., 2011). HIV-positive women may require multiple doses of metronidazole because of changes in vaginal ecology, interference of impaired immunity with single-dose treatment, and interaction of antiretroviral drugs with metronidazole (Kissinger and Adamski, 2013). In men's case, *T. vaginalis* infections are also associated with chronic prostatitis leading to aggressive prostate cancers, as observed from increased Prostate-Specific Antigen levels (Langston et al., 2019; Suitcliffe et al., 2006).

Even though the single metronidazole therapy has a failure rate of only 10%, these figures are significant due to the large number of patients suffering from trichomoniasis. Although oral 5-nitroimidazoles such as metronidazole and tinidazole exhibit high cure rates, trichomonas infection can still be highly persistent and recurrent (Dunne et al., 2003; Seña et al., 2014). One of the major reasons for this is the drug resistance to metronidazole or cross-resistance to other 5-nitroimidazoles or, in some cases, multiple drug resistance (Dunne et al., 2003). Additionally, in certain cases, metronidazole-associated allergy may cause urticaria, facial edema, and anaphylactic shock. It may result in therapy failure as well (Mehriardestani et al., 2017). Drug resistance to metronidazole or the whole 5-nitroimidazole family is fairly common, which eventually exposes the lack of drugs available in the armamentarium to treat trichomonas infection. Given the population density that suffers from trichomonas infection and lack of drugs, there is an urgent need to discover safe and efficacious drugs to treat trichomonosis.

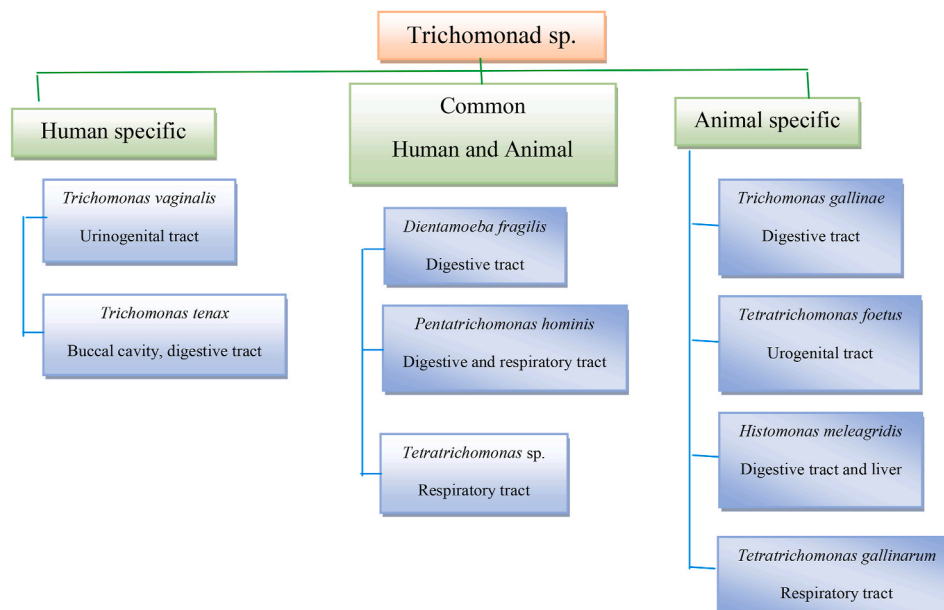On a different but serious note, trichomonads are evolving and losing

Fig. 1. Trichomonas species and their infection site in the host.

strict host specificity; *T. vaginalis*-like isolates from cases of epidemic avian trichomoniasis exemplify the importance to create awareness of potential human-to-bird transfer and evolution and origins of these pathogens (Maritz et al., 2014). Cross infection of parasites between pigs and cattle has also been observed (Miller et al., 2017). Trichomonad parasites, which were known to infect animals, are now causing infection to humans as well. Although rare, human tritrichomoniasis caused by *T. foetus* has been reported as opportunistic infections in immuno-compromised or immunosuppressed individuals (Suzuki et al., 2016). *T. foetus* is also found in the stomach, caecum, and nasal cavity of pigs without apparent clinical significance (Mueller et al., 2015).

This report's objective was to collate the literature on activity against *T. vaginalis* of isolated natural products and whole plant products. Several reviews have analyzed medicinal plants' efficacy and safety, their extracts, or isolated plant constituents for treating *T. vaginalis* infection. However, the results of systematic reviews are not consistent, and the methodological quality of systematic reviews is unknown. No overview focused on the efficacy and safety of several plant species. This study aimed to summarize the evidence from reviews and systematic reviews that evaluated the effectiveness of various plants and their isolated constituents in treating trichomonas infection and critically assessing the evidence's quality.

2. Materials and methods

2.1. Review question

This systematic review aimed to look at the evidence across the scientific literature for the efficacy of various plants to treat *Trichomonas vaginalis* infection.

2.2. Search strategy

The protocol of this qualitative synthesis of the current literature is performed according to the recommendations of the PRISMA Statement [Moher et al., 2015] with the relative flow diagram shown in Fig. 1. US National Library of Medicine (PubMed), ScienceDirect® and Scopus® trademark of Elsevier, Google Scholar, Wiley Interscience, Taylor, and Francis Group were searched combining the terms ("*Trichomonas vaginalis* infection" or "trichomoniasis" or "anti-trichomonal") and ("natural treatment" or "plant therapy" or "cure" or "plant extract" or "medicinal plant" or "herb" or "herbal medicine"). We chose all the full-text articles published in the English language only evaluating anti- *T. vaginalis* medicinal plants using the keywords mentioned above. Several articles were found to end tracking citations from other publications or directly access the journals' website.

2.3. Quality assessment and article selection

All the articles identified in the databases mentioned above were evaluated independently by two reviewers. After reviewing the titles, abstracts, and full texts of the articles, unrelated studies were excluded from the review. The remaining articles were investigated using quality assessment checklists.

2.4. Inclusion criteria

All the study articles evaluating the *in-vitro* and *in-vivo* effects of medicinal herbs on *T. vaginalis*, published between 2000 and 2020.

2.5. Exclusion criteria

All the articles with studies shown outside the determined period and articles on parasite species other than *Trichomonas vaginalis*.

2.6. Data extraction

Essential data with the scientific name of plants, type of herbal extracts, used parts of the plants, extract concentrations, and killing or growth-inhibitory effects were obtained from the selected articles and recorded.

3. Results and discussion

3.1. Search results

At first, nine possibly relevant records were retrieved. After screening titles and abstracts, 20 records were excluded, and the remaining 79 records were considered potentially eligible for full-text screening. Lastly, 55 reviews were included in this overview (Fig. 2).

Drugs from medicinal plants have proven to be effective, cost-effective, well-tolerated, and with fewer side effects than synthetic drugs. It has prompted scientists globally to screen medicinal plants for the effective treatment of trichomoniasis. Medicinal plants are a huge source of compounds with a diverse structural range. Some of the plants/phytopharmaceuticals are reported to possess anti-*Trichomonas* activity. The largest number of plants reported showing anti-*Trichomonas* activity belong to the *Asteracea, Lamiaceae,* and *Myrtaceae* families of plants. The majority of active phytopharmaceuticals were found to fall under the categories of terpenes, β-glycosides, saponins, essential oils, and alkaloids (Mehriardestani et al., 2017).

3.2. Description of included systematic reviews on isolated compounds

3.2.1. Isolated natural product- alkaloid berberine

Berberine is a nonbasic and quaternary benzylisoquinoline alkaloid, isolated from plants with a long history of medicinal use in the traditional medicine system.
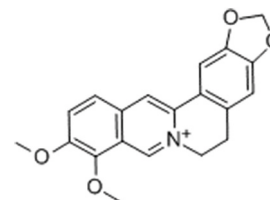
Berberine has been isolated from various plant families (Kaneda et al., 1990; Neag et al., 2018), as given in Table 1.

However, the genus *Berberis* is the best potential berberine source (Imenshahidi and Hosseinzadeh, 2016).

Berberine is often administered in a salt form for several clinical applications like anti-bacterial, anti-fungal, anti-inflammatory, anti-malarial, antioxidant, and analgesic activities (Pund et al., 2014a). Exposure of *T. vaginalis* to berberine sulfate caused deformity in the shape of the organism. Autophagic vacuoles were formed and increased in number, leading to the plasma membrane's rupture, thus exhibiting anti-trichomonal activity (Kaneda et al., 1990). Berberine hydrochloride intravaginal therapy had shown significant improvement in bacterial vaginosis patients' clinical symptoms as observed from a reduction in oxidative stress in vaginal discharge and apoptosis of vaginal epithelial cells. The activities of superoxide dismutase and catalase were noted as reduced levels of malondialdehyde and hydrogen peroxide (Ma et al., 2019). However, the exact infection cause of vaginitis was not identified in these patients. Soffar et al. (2001) suggested berberine sulfate as a safe alternative to metronidazole, having comparable potency and applicability in metronidazole resistant cases.

Considering berberine as one of the important constituents of plant *Argemone mexicana*, very recently, Elizondo-Luevano et al. (2020) compared the anti-trichomonal effect of methanolic extract of leaves and stem of the plant with pristine berberine and metronidazole as a
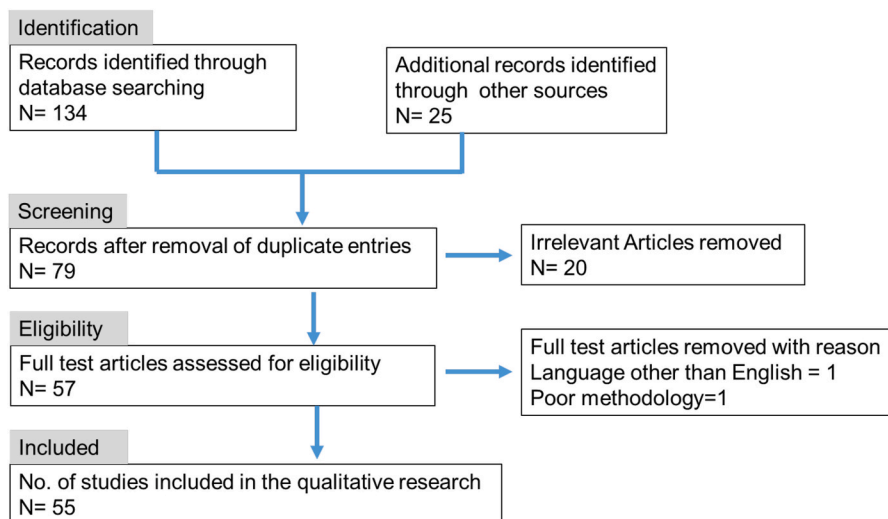
**Fig. 2.** Flow chart describing the study design process.

Table 1
Various plant families and species containing berberine alkaloids, having the potential for antitrichomonal activity.

Plant family	Genera	Examples of plant species
Annonaceae	*Annickia, Coelocline, Rollinia, Xylopia,*	*A. chlorantha (Oliv.) Setten and Maas, A.pilosa (Exell) Setten and Maas, R. pilocarpa Loefgr., C. polycarpa, R. mucosa (Jacq.) Baill., X. macrocarpa, X. polycarpa (DC.) Oliv.*
Berberidaceae	*Berberis, Caulophyllum, Jeffersonia, Mahonia, Nandina, Sinopodophyllum*	*B. aristata DC., B. aquifolium Pursh, B. heterophylla Juss. ex Poir., B. beaniana C.K. Schneid., B. asiatica Roxb., B. croatica Horvat, B. thunbergii DC., B. vulgaris L., B. pseudumbellata R. Parker*
Menispermaceae	*Coscinium, Tinospora*	*C. fenestratum Colebr., T. cordifolia (Willd.) Miers*
Papaveraceae	*Argemone, Bocconia, Chelidonium, Corydalis, Eschscholzia, Glaucium, Hunnemannia, Macleaya, Papaver,* and *Sanguinaria*	*A. alba F. Lestib., A. Mexicana L., A. hispidu, A. munita Durand and Hilg., B. frutescens L., Chelidonium majus L., Corydalis solida (L.) Clairv., Corydalis turtschaninovii Besser, E. californica Cham., G. corniculatum (L.) Rudolph, P. rhoeas L.,*
Ranunculaceae	*Coptis, Hydrastis,* and *Xanthorhiza*	*C. chinensis Franch., C. japonica (Thunb.) Makino, C. rhizome, H. Canadensis L., X. simplicissima Marshall*
Rutaceae	*Evodia, Phellodendron,* and *Zanthoxyllum*	*E. meliaefolia, P. amurense Rupr., P. chinense C.K. Schneid., P. lavallei Dode, Z. quinduense*

standard control. The IC_{50} values of stem and leaf extracts of *Argemone mexicana* against *T. vaginalis* were 70.8 and 67.2 µg/mL, respectively. Although the values observed were not comparable to that of metronidazole, IC_{50} 0.1 µg/mL, they were comparable to pure berberine of IC_{50} 40.7 µg/mL.

3.2.2. Isolated natural product- polyphenol curcumin

Curcumin is a polyphenol extracted from the Indian spice turmeric, *Curcuma longa*, a rhizomatous perennial plant.
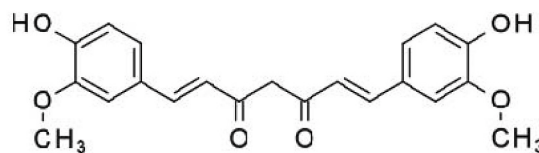

In Indian food, it is traditionally used for food coloring and has been investigated for its biological research outcomes over the past years (Wachter et al., 2014). Its diverse therapeutic activity encompasses anti-inflammatory, anti-cancer, antioxidant, immunomodulatory action, and anti-infective (Pund et al., 2016). Curcumin is an effective anti-infective agent against various parasites, e.g., *Giardia lamblia* (Perez-Arriaga et al., 2006)*, Eimeria tenella* sporozoites (Khalafalla et al., 2011)*, Plasmodium* spp. (Mimche and Taramelli 2011)*, Cryptosporidium parvum* (Shahiduzzaman et al., 2009)*, Leishmania amazonensis,* and *L. mexicana* (Koide et al., 2002)*, Trypanosoma brucei* (Changtam et al., 2010)*, Schistosoma mansoni* (Allam, 2009), and HIV (Jordan and Drew 1996). Various studies suggest the *in-vitro* therapeutic utility and efficacy of curcumin in trichomoniasis. Wacther et al. (2014) studied the effectiveness of curcumin in various concentrations against *T. vaginalis.* The group also studied the efficacy of curcumin in metronidazole-resistant strains of *T. vaginalis* (Wachter et al., 2014). Curcumin eradicated 100% *T. vaginalis* cells at the concentration of 400 µg/mL in 24 h. The EC_{50} ranged from 73.0 to 105.8 µg/mL, while the EC_{90} from 164.9 to 216.3 µg/mL. Although in comparison to metronidazole, curcumin was found to be effective at a higher concentration, curcumin is well tolerated without any side effects in contrast to metronidazole. The safety of curcumin is well established and demonstrated. However, these concentrations are very high, even to a natural compound that the authors presented without toxicity. As demonstrated by Cos et al. (2006), endpoints, such as IC50, higher than 100 µg/mL for extract and 25 µM for isolated compounds, should not be considered promising.

A systematic meta-analysis was carried out to study the safety of curcumin (Chainani, 2003). This meta-analysis included various research articles, book chapters, *in-vitro* studies, human trials from 1966 to 2003. The metanalysis reported that curcumin is a well-tolerated compound and is safe in at least six human clinical trials. Moreover, curcumin can be administered topically, which will further reduce the likelihood of side effects associated with metronidazole's oral administration. Thus, curcumin holds a promise as a potential anti-trichomoniasis candidate of plant origin. However, the poor aqueous solubility of curcumin limits its widespread therapeutic

utilization. To overcome the poor aqueous solubility (Carapina da Silva et al., 2019) synthesized 21 monocarbonyl analogs of curcumin that possessed anti-protozoal activity. Out of these 21 compounds, three derivates, 1,5-diphenylpenta-1,4-dien-3-one, 1,5-bis(2-chlorophenyl) penta-1,4-dien-3-one, and 2,6-bis(2-chlorobenzylidene) cyclohexanone, were found to have comparable anti-trichomoniasis activity as that of metronidazole. All the three derivates eradicated *T. vaginalis in vitro*. Additionally, 1,5-bis(2-chlorophenyl) penta-1,4-dien-3-one was found to have higher selectivity as well (Carapina da Silva et al., 2019). However, two reports suggest a contradictory effect of curcumin on *T. vaginalis* (Muelas-Serrano et al., 2000; Raether and Seidenath, 1984).

### 3.2.3. Isolated natural product – polyphenol resveratrol

Resveratrol is a dietary non-flavonoid polyphenolic phytoalexin, present in grapes' skin and exhibits a broad range of biological activities that potentially underlie its antioxidant anti-inflammatory, and anti-carcinogenic efficacy (Pund et al., 2014b).
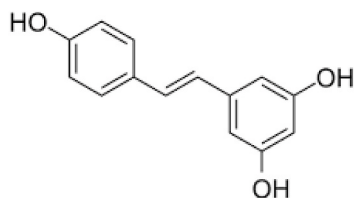
In a mechanistic study, Mallo et al. (2013) explored the *in-vitro* antitrichomonal effect and mechanism of action of resveratrol on *T. vaginalis* trophozoites. At concentrations between 25 and 100 μM, resveratrol inhibited the growth of *T. vaginalis* trophozoites. The lower dose of 25 μM exerted a cytostatic effect, whereas higher doses exerted killing effects. Resveratrol, at a high dose, upregulated gene expression of the hydrogenosomal enzyme pyruvate-ferredoxin oxidoreductase. At doses lower than 100 μM but more than 50 μM, overexpression of heat shock protein 70, a protective protein found in the hydrogenosome of *T. vaginalis,* was observed. The results demonstrated that resveratrol exerts its deleterious effects as anti-trichomonas via hydrogenosomal dysfunction and metabolic alteration (Mallo et al., 2013).

3.2.4. Isolated natural product- anthraquinone emodin

Emodin, a derived anthraquinone, is extracted from roots and barks of various plants like *Rheum palmatum, Aloe vera, Polygonum multiflorum,* and *Polygonum cuspidatum.* Emodin shows fundamental therapeutic properties, such as antioxidant, anti-inflammatory, anti-bacterial, and anti-viral (Cui et al., 2020).
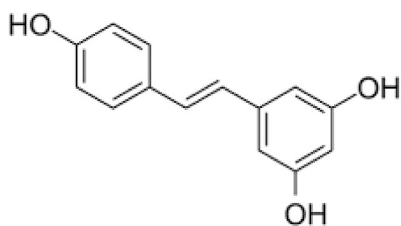
Wang et al. (1993) studied *in-vivo* efficacy of emodin in two types of experimental infections, namely that caused by subcutaneous injection of *T. vaginalis* in BALB/c mice and the one due to intravaginal inoculation of the parasites in mice. The appearance of subcutaneous abscess induced by injection of trichomonads was delayed by emodin treatment. An oral administration also cured the trichomonal abscesses, indicating good absorption of emodin anthraquinone. Oral treatment with emodin significantly decreased the number of trichomonads in the vagina as well. This activity's criteria were determined according to living trichomonads in isolated vagina obtained from the treated mice.

3.2.5. Isolated natural product-flavonoid quercetin and quercitrin
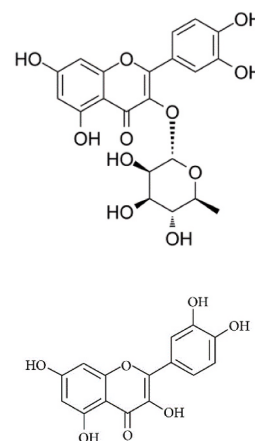
Omisore et al. (2005) studied the anti-trichomonas activity of *Dorstenia barteri*, and *Dorstenia convexa* extracts, and some isolated components from *D. barteri* against *T. gallinarum* and compared with quercetin and quercitrin. Quercetin was found to be most potent, followed by quercitrin. Minimum lethal concentrations were found to be 0.121., 0.244, 0.244, 0.73, and 0.98 μg/mL for quercetin, quercitrin, bartericin B, bartericin A, and stigmasterol, respectively, and for 6, 8-diprenyleridictyol, isobavachalcone, and dorsmanin F, similar activity was noted i.e., 31.25 μg/mL. In the same study, researchers studied minimum lethal concentrations for the extract of leaves and twigs of both the *Dorstenia* species. Extracts were prepared using a 1:1 organic solvent mixture of dichloromethane and methanol. At 24 h exposure, minimum lethal concentrations were found to be 15.625 and 15.625 μ/mL for extracts of leaves and twig of *D. barteri,* whereas leaf and twig extract of *D. convexa* exhibited much higher minimum lethal concentrations as 125 and 437.5 μg/mL, respectively.

Naemi et al. (2014) have reported anti-trichomonas activity of the plant *Rheum ribes* L. extracts*,* which contains quercetin, and derivatives of quercetin like 5-desoxyquercetin, quercetin 3-*O*-rhamnoside, quercetin 3-*O*-galactoside, and quercetin 3-*O*-rutinoside. Aqueous and organic extracts of flowers, leaves, and stem showed *in-vitro* anti-trichomonal activity against *T. vaginalis* at a 125 μg/mL concentration.

Brandelli et al. (2013) showed remarkable anti-trichomonas activity of ten different plants traditionally used by the Mbyá-Guarani indigenous group in Brazil. Researchers studied extracts of leaves of *Aloe arborescens* Mill., aerial parts of *Bidens pilosa* L., aerial parts of *Rhipsalis baccifera* (Sol.) Stearn, barks of *Luehea divaricate* Mart., roots of *Trichilia* sp., leaves of *Campomanesia xanthocarpa* Mart. ex O. Berg, leaves of *Coix lacryma-jobi* Lin., leaves of *Citrus limonium*, leaves of *Citrus reticulate* Blanco, and *Verbena* sp leaves. The anti-*Trichomonas vaginalis* activity was attributed to flavonoids, namely quercetin, quercitrin, myricetin, and rutin.

3.2.6. Isolated natural product- alkaloid lycorine

Lycorine is the most common Amaryllidaceae phenanthridine ring-type alkaloid present in Amaryllidaceae family plants and has been considered a promising anti-proliferative agent.
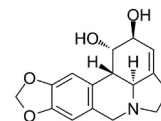
This natural alkaloid has immense therapeutic potential as it has shown high specificity in a very low concentration against several cancers both *in vivo* and *in vitro* and against various drug-resistant cancer cells (Roy et al., 2018). Recently, lycorine showed anti-viral activity against the Zika virus *in vitro* and *in vivo* (Chen et al., 2020). Giordani et al. (2011) studied the cytotoxicity of lycorine against *T. vaginalis*

covering a very broad range of concentrations from 2.5 to 1000 μM. Cell cycle analysis and observations of ultrastructural alterations in the parasitic cell were analyzed by flow cytometry and transmission electron microscopy, respectively, after treatment with 250 μM lycorine for 6 and 24 h. Significant changes occurred, including disruption in the trophozoite's original shape and a few depressions on its surface. Although apoptotic blebs were not seen, the nucleus looked abnormal in shape, e.g., elongated or split. Several cytoplasmic vacuoles resembling an autophagic process were observed. Randomly spread hydrogenosomes were also seen within the parasite cells. Fragments of the endoplasmic reticulum were seen in close contact with abnormal hydrogenosomes, indicative of autophagy. Lycorine arrested the T. vaginalis cell cycle at the G2/M phase of the cell cycle, although no apoptosis hallmarks, such as apoptotic bodies, were observed. Consequently, the underlying mechanism of action fails to fulfill the criteria for apoptosis completely. However, some similarities to paraptotic cell death were observed (Giordani et al., 2011).

3.2.7. Isolated natural product alkaloid candimine

Candimine, an alkaloid from the bulbs of *Hippeastrum morelianum*, was found to be cytotoxic for the parasite *T. vaginalis* (Giordani et al., 2010a). An ultrastructural study of the parasite in the presence of candimine revealed striking morphological alterations, although the cell membrane remained intact in all parasites observed. After treatment with 1 for 6 and 24 h, modifications in size, shape, and intracellular localization of hydrogenosomes. The hydrogenosomes presented signs of autophagy. Besides, candimine caused arrest of the *T. vaginalis* cell cycle, altered morphologic and ultrastructural features, but did not induce the formation of apoptotic bodies, left the cell membrane intact, and did not cause exposure of phosphatidylserine residues nor DNA fragmentation. Thus, candimine-induced cell death in *T. vaginalis* fails to fulfill apoptosis criteria and instead exhibits features like paraptosis.

The same group (Giordani et al., 2010b) studied the effects of both lycorine and candimine alkaloids on the *T. vaginalis* NTPDase and ecto-5′-nucleotidase activities. When both alkaloids were added directly to the reaction mixture, no effect on ATP, ADP, or AMP hydrolysis could be observed. In contrast, *T. vaginalis* NTPDase, and ecto-5′-nucleotidase activities were significantly inhibited by candimine and lycorine after 24 h treatment. Expressive inhibition by lycorine and candimine on this enzymatic cascade in *T. vaginalis* modulates the extracellular ATP and adenosine levels. The accumulation of extracellular nucleotides, mainly the cytotoxic and proinflammatory ATP due to alkaloids inhibition, plus the ATP originated from host cells, associated to reduced concentrations of the immune-modulatory adenosine, might contribute to an increased susceptibility of *T. vaginalis* to host immune response in the presence of lycorine and candimine (Giordani et al., 2010b). The fact that ecto-nucleotidases have an external location are implicated in parasite survival and are widely distributed among different eukaryotic human pathogens make them interesting targets for the development of antimicrobial agents. This study claimed a new pharmacological potential of two Amaryllidaceae alkaloids, lycorine, and candimine against *T. vaginalis.*

3.2.8. Isolated natural product- monoterpenoid geraniol

Dai et al. (2016) studied the essential oil of *Amomum tsao-ko* Crevost and Lemarié, which belongs to the family Zingiberaceae, and its primary constituent geraniol, for anti-*Trichomonas vaginalis* activity.
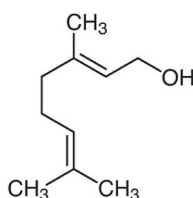
It is a spice used in Chinese cuisine and is also part of the Chinese Traditional Medicine. *A. tsai-ko* is effective for the treatment of stomach disorders and throat infection (and exhibits a wide range of biological activities, such as antimicrobial, antioxidant, and cytotoxic activities, as well as lipid-lowering effects). Geraniol is a widely used fragrance ingredient and is the main component of *A. tsao-ko* essential oil, constituting approximately 14%. Geraniol has antimicrobial activity, insecticidal, and antitumor activity (Kim et al., 2011). Both *A. tsao-ko* essential oil and geraniol showed *in-vitro* anti-*T. vaginalis* activity against two different parasite isolates. *A. tsao-ko* essential oil was found to be 4-8-fold more effective than that of geraniol; however, it was 10-fold less active than metronidazole, 20-fold less than ornidazole Dai et al. (2016) further explored the mechanism of activity of *A. tsao-ko* essential oil and geraniol against *T. vaginalis* using direct visualization with transmission electron microscopy. Several morphological changes were observed in the treated cells, such as many vacuoles, disappearance of ribosomes, dilation of rough endoplasmic reticulum, and dissolving nuclei. The leakage of cytoplasmic contents was also observed.

3.2.9. Isolated plant compounds- β-glycosides

Arthan et al. (2008) isolated various β-glucosides and their corresponding aglycone part from various Thai plants (Table 2).

Among these 11 compounds tested, kingiside, gonocaryoside A, plumieride coumarate glucoside, and torvoside A were the most potent anti-trichomonal with MIC of 6.25 μM. Although these compounds were 125-fold less effective than metronidazole (MIC 0.05 μM), they were found to be safe and not toxic on Vero, KB, and MCF-7 cell lines at the concentrations tested.

3.2.10. Isolated plant product- diterpenoids and triterpenoids

Loyola et al. (2001) isolated diterpenoids, namely 9,12-cyclomulin-13-ol or 13β-hydroxyazorellane,13α-hydroxyazorellane, azorellanol, mulinolic acid, and mulin-11,13-dien-20-oic acid from the aerial parts of *Azorella yareta* Hauman. All the compounds showed anti-trichomonal activity *in vitro*; however, it was very low, as observed from very high LC_{50} values, 40 ± 120 μM, which is almost ten times higher than of metronidazole ($LC_{50} = 6.6$ μM).

Betulinic acid and ursolic acid were isolated from barks of *Platanus acerifolia*, and *Malus domestica* peels, respectively (Innocente et al., 2014), and semisynthetic derivatives of both were prepared. Compounds obtained from betulinic acid presented better activity than those from ursolic acid. Piperazine derivatized from betulinic acid presented a minimum inhibitory concentration of 91.2 μM, revealing these compounds' high potential as trichomonacidal agents.

A pentacyclic triterpenoid, hederagenin isolated from *Cussonia holstii* is a traditional African medicinal product that was found to display antitrichomonal activity with an IC_{50} of 2.8 μM (He et al., 2003).

3.2.11. Isolated plant product- saponins

Rocha et al. (2012) successfully demonstrated saponins' activity

Table 2
β-Glucosides and their corresponding aglycone part from various Thai plants isolated for anti-trichomonas activity (Arthan et al., 2008).

Plant and part of the plant	β-glucosides	Aglycone
Thai rosewood seeds	dalcochinin-8′-*O*-β-D-glucoside	Dalcochinin
Solanum torvum Sw. fruits	torvoside A, torvoside H	26-degluco-torvoside A 26-degluco-torvoside H
Plumeria obtusa L. flowers	Plumieride coumarate glucoside	13-*O*-coumarylplumieride aglycone
Gonocaryum subrostratum Pierre leaves	Gonocaryoside A, kingiside	–
Ligustrum confusum Decne. leaves	Ligustaloside A	–

from *Quillaja*, *Passiflora*, and *Ilex* species on *T. vaginalis*. Saponins were extracted from the leaves of *Passiflora alata* Curtis (passion fruit), *Quillaja brasiliensis* Mart. (soap tree), and *Ilex paraguariensis* A. St.-Hil. (maté).

Saponins from *Sapindus mukorossi* showed anti-*Trichomonas* activity at a 10-fold lower concentration than required for effective spermicidal activity against human spermatozoa, thus suggesting the potential of these saponins for the development of a safe spermicidal and microbicide for human use (Tiwari et al., 2008). Jain et al. (2014) combined a synthetic spermicide with a natural saponin trichomonacide for safe, prophylactic contraception. This study's anti-trichomonal saponins were a fully standardized mixture of six sapindosides (sapindosides A, B, C, D, and mukurozisaponins E1 and Y1), with sapindoside B as the major constituent. They were isolated from the fruit pericarp of *Sapindus mukorossi* Gaerth (family Sapindaceae) by *n*-butanol extraction of its ethanolic extract. The combination of saponin with synthetic spermicide resulted in lowering the anti-trichomonal MIC of synthetic spermicide from 285 μg/mL to 32.8285 μg/mL.

Damke et al. (2013) evaluated the anti-trichomonas activity of a purified sample of saponins and hydroalcoholic and butanolic extract of dried pericarps of *Sapindus fruits saponaria*. MIC for saponins was four-fold less than for hydroalcoholic extract and two-fold less than butanolic extract.

3.2.12. Aromatic plant extracts and essential oils

Essential oils are the natural, volatile, aromatic, complex secondary metabolites produced in aromatic plants and are characterized by strong aroma (Bakkali et al., 2008). Such aromatic plants generally grow in warm and tropical countries. Essential oils are known to possess medicinal properties, e.g., virucidal, fungicidal, antiseptic, and bactericidal. Some of the essential oils like those from *Mentha piperita* (peppermint), *Salvia officinalis* (Yousefi et al., 2013) *Zataria multiflora* (Mahboubi, 2018), and *Lavandula angustifolia* (lavender) (Moon et al., 2006) possess anti-trichomonal properties. All four belong to the aromatic plants of the Lamiaceae or Labiatae family. Hydro-alcoholic extracts of *M. piperita* and *S. officinalis* were found to have anti-trichomonal activity comparable to metronidazole. However, *S. officinalis* was found to have higher potency than *M. piperita* (Yousefi et al., 2013). From the same family, *Mentha crispa* has been evaluated for its efficacy as anti-trichomonal activity in a clinical trial (Moraes et al., 2012). In a randomized, double-blind, and controlled clinical trial, 60 female patients were administered orally with 90% of the patients administered with *M. crispa* was found to have recovered from trichomoniasis while 96.6% recovered using secnidazole [statistically not significantly different ($p = 0.6120$)]. However, the secnidazole group's adverse events were statistically significantly higher than the *M. crispa* group ($p = 0.0006$). *Mentha crispa* exhibited comparable therapeutic efficacy as that of secnidazole with significantly lower side effects (Moraes et al., 2012). In another *in-vitro* study, the efficiency of *Mentha longifolia* extract was compared with that of aqueous extract of *Pimpinella anisum* against a clinical strain of *T. vaginalis* (Niyyati et al., 2015). Aqueous extract of *Mentha longifolia* reduced the number of live *T. vaginalis* in a time- and concentration-dependent manner and exhibited a MIC of 250 μg/mL and 200 μg/mL at the end of 24 h and 48 h, respectively. In contrast, an extract of *Pimpinella anisum* inhibited only 73.7% and 86.9% of live parasites at a 500 μg/mL concentration at the end of 24 h and 48 h, respectively. *Mentha longifolia* extract seems to possess higher potency towards *T. vaginalis* than *Pimpinella anisum* (Niyyati et al., 2015). Further, Aslani et al. (2019) evaluated a vaginal cream for anti-trichomonal activity containing a combination of *Eucalyptus camaldulensis*, *Viola odorata,* and *Mentha piperita* extracts. A cream preparation containing 2.5 mg/mL of *E. camaldulensis*, 0.06 mg/mL of *V. odorata*, and 1 mg/mL of *M. piperita* was found to inhibit 100% growth of *T. vaginalis* at the end of 24 h. *Z. multiflora* is another flowering plant of the Lamiaceae family that has been reported to possess anti-trichomonal properties. *Z. multiflora* contains oxygenated

sesquiterpenes (1.6–5.7%), monoterpene hydrocarbons (3.6–24.5%), thymol, carvacrol and linalool, and *p*-cymene (Mahboubi, 2018). *Z. multiflora* essential oil at a concentration of 0.1–0.0004% was found to inhibit clinical isolates of *T. vaginalis* at 1 h (Abdollahy et al., 2004). Efficiency of *Z. multiflora* vaginal cream, 0.1%, as compared with metronidazole in 54 non-pregnant women. 88.9% of patients were treated in the metronidazole group than 51.9% of the *Z. multiflora* group. A group of 48.1% of patients treated with metronidazole showed adverse effects compared to none of the patients treated with *Z. multiflora*. The authors concluded that though the clinical success was low in *Z. multiflora*-treated patients, still the patients of this group exhibited greater improvement in clinical symptoms (Abdali et al., 2015).

Essential oils from *Lavandula angustifolia* and *Lavandula intermedia* were evaluated for their anti-trichomonal properties (Moon et al., 2006). It was found that both the lavender essential oils can eradicate *T. vaginalis*, even at a concentration below 1% (Moon et al., 2006).

Black seed or black cumin or *Nigella sativa* L., which belongs to the Ranunculaceae, has a widespread culinary and traditional medicinal usage in the Mediterranean and Indian subcontinent. *Nigella sativa* L. seeds have been reported to possess antioxidant, anti-inflammatory, anti-fungal, anti-infective, and anti-parasitic properties. The effect of aqueous extract of *N. sativa* was compared with metronidazole on the growth and motility of *T. vaginalis* (Tonkal, 2009). Aqueous extract of *N. sativa* was found to possess a remarkable effect on the growth of *T. vaginalis*. To further elucidate the mechanism and understand the effect of fatty acids of *N. sativa*, Mahmoud et al. (2016) undertook a study to compare the efficacy of *N. sativa* oil, aqueous extract, and alcoholic extract of *N. sativa* on *T. vaginalis*. It was found that *N. sativa* oil at a concentration of 2 mg/mL, and alcoholic extract at a concentration of 10 mg/mL, exhibited similar activity as that of metronidazole (50 μg/mL) after 24 h. The aqueous extract was found to have less effect on *T. vaginalis*. This difference in the activity was attributed to the fatty acids present in *N. sativa* oil and those extracted in the alcoholic extract of *N. sativa*. The lowest anti-trichomonal activity of aqueous extract of *N. sativa* was ascribed to the absence of major fatty acids, viz., oleic acid, and linoleic acid, in the aqueous extract. Remarkably, the fatty acids tridecanoic acid and palmitoleic acid were found only in the oil of *N. sativa*. The authors proposed a possible mechanism of action that suggests the cell membrane of *T. vaginalis* as the main target. It is presumed that fatty acids in the oil and alcoholic extract may interact with the cell membranes, resulting in transient or permanent variable size pores. This formation of pores results in leakage, reduced nutrient uptake, or cellular respiration inhibition in *T. vaginalis* (Mahmoud et al., 2016). Thus, oil and alcoholic extract of *N. sativa* are potentially bioactive against *T. vaginalis*.

Rosemary, an age-old globally used herb known to strengthen brain and memory activity, was found to possess antitrichomonal activity when the extract, rather than an essential oil, was used (Saeidi et al., 2019). Rosemary essential oil essentially contains camphor, 1,8-cineole, verbenone, borneol, and α-pinene. In the same line, essential oil from *Artemisia sieberi* was effective both *in vitro* and *in vivo* in an infected pigeon model against *Trichomonas gallinae* (Youssefi et al., 2017). *Pistacia lentiscus mastic* and *Ocimum basilicum* oil are known for their anti-bacterial, anti-fungal, anti-viral, and anti-protozoal effects were compared for anti-trichomonal activity (Ezz Eldin and Badawy, 2015). Amongst the two, *Ocimum basilicum* oil was found to be more potent as compared to *Pistacia lentiscus mastic* oil. The minimum lethal dose of the *Pistacia lentiscus mastic* oil was found to be 15 mg/mL after 24 h incubation, 10 mg/mL after 48 h, and 5 mg/mL after 96 h, while the minimal lethal concentration of *O. basilicum oil* was 30 μg/mL after 24 h incubation, 20 μg/mL after 48 h and 10 μg/mL after 96 h. The essential oils disrupt the cellular membrane or cause extensive vacuolization of cytoplasm, resulting in eradicating the parasite *T. vaginalis* (Mehriardestani et al., 2017). These essential oils need to be screened and clinically evaluated further for their anti-trichomonal activity. van Vuuren

and Naidoo (2010) also tested essential oils from *Croton gratissimus* and *Tarchonanthus*. However, they were found to be ineffective against *T vaginalis*.

3.3. Extracts of the whole plant or plant parts

3.3.1. Eucalyptus

One such plant that has been found to possess potential anti-trichomonas activity is *Eucalyptus camaldulensis*; growth-inhibitory activity against *T. vaginalis* has been reported for the extract of *E. camaldulensis* (Mehriardestani et al., 2017). *E. camaldulensis* or the River Red Gun tree belongs to the genus *Eucalyptus* and family Myrtaceae. *Eucalyptus* trees are well known for their adaptability and ability to grow fast. Consequently, *Eucalyptus* is one of the widely grown plants globally. Of the 800 species in the *Eucalyptus* genus, *E. camaldulensis* is one the most grown. Being a native of Australia and Tasmania, *E. camaldulensis* is a widely used medicinal plant in Aboriginal society and Australia's traditional medicinal system (Aleksic Sabo and Knezevic, 2019). The medicinal application of *E. camaldulensis* ranges from treating gastrointestinal disorders, e.g., diarrhea, colic, and dysentery, to respiratory diseases such as colds, coughs, laryngitis, and sore throat (Aleksic Sabo and Knezevic, 2019). It has also been traditionally used to arrest bleeding and joint pain. However, it is used as a folk medicine globally. Smoked leaves of *Eucalyptus* are used to alleviate respiratory issues in Sudan, while its decoction is used for treating coughs and colds in Zimbabwe. Nigerian people use its stem for oral hygiene. The medicinal properties of *Eucalyptus* are attributed to its chemical constituents. *Eucalyptus* is rich in essential oils ~0.4%, primarily 1,8-cineole (~77%). Other major constituents are cuminal, phellandrene, aromadendrene (or aromadendral), valerylaldehyde, geraniol, cymene, and phellandral. The leaves also contain a considerable quantity of tannins (Ghasemian et al., 2019). *E. camaldulensis* has been proven to possess a diverse range of medicinal properties such as anti-bacterial, anti-fungal, anti-viral, and anti-protozoal. The first reported anti-trichomonas activity of *E. camaldulensis* was by Mahdi and co-workers (Mahdi et al., 2006). This group reported that an aqueous extract of *E. camaldulensis* leaves (at a concentration of 500 mg/mL) at pH 5.35 resulted in eradicating *T. vaginalis* at the end of 24 h (Mahdi et al., 2006). Later, a couple of other groups further confirmed this finding in independent research. Hassani et al. (2013) evaluated five different types of *E. camaldulensis* extract on *T. vaginalis* (Hassani et al., 2013). The group evaluated total extract, diethyl ether, chloroform, ethyl acetate, and water fractions of *E. camaldulensis in vitro*. The growth-inhibitory activity was found with all the extracts. The potency of all the extracts for the growth inhibition of *T. vaginalis* can be ranked as ethyl acetate extract > crude extract > diethyl ether extract > water extract. Ethyl acetate extract exhibited 100% growth inhibition of *T. vaginalis* at a concentration of 12.5 mg/mL at the end of 24 h, while the water extract exhibited 80% growth inhibition at a concentration of 50 mg/mL at the end of 24 h (Hassani et al., 2013). Around the same time, the findings of another group further strengthened these results. Youse et al. (2012) compared alcoholic and water extracts of *Echinophora platyloba, Stachys lavandulifolia*, and *E. camaldulensis* for *in-vitro* activity against *T. vaginalis* (Youse et al., 2012). Out of the three plant extracts, only the extracts of *E. camaldulensis* exhibited *in-vitro* activity against *T. vaginalis*. Based on these findings, Aslani et al. (2019) developed a vaginal cream containing extract of *E. camaldulensis* along with extract of *Viola odorata* and *Mentha piperita* (Aslani et al., 2019). A cream formulation containing 2.5 mg/mL of *E. camaldulensis*, 0.06 mg/mL of *V. odorata*, and 1 mg/mL of *M. piperita* was found to inhibit 100% growth of *T. vaginalis* at the end of 24 h. While the formulation containing 1.25 mg/mL of *E. camaldulensis*, 0.03 mg/mL of *V. odorata*, and 0.5 mg/mL of *M. piperita* exhibited only 92% of growth inhibition at the end (Ghasemian et al., 2019) of 24 h.

3.3.2. American plants

In a very exhaustive experiment, Muelas-Serrano et al. (2000) systematically explored the anti-trichomonas activity of various aqueous and organic extracts of different parts of more than 40 South American plants belonging to 20 different families known for their immunomodulatory, anti-inflammatory, and anti-protozoal effects (Table 3). Cytocidal and cytostatic activities against *T. vaginalis* strain JH31A no. 4 were evaluated concerning control cultures. *Mikania cordifolia* (Linnaeus f.) Willdenow and *Neurolaena lobata* (L.) R. Br. from the Asteraceae and *Scutia buxifolia* Reiss from the Ramnaceae were the most active extracts amongst all the plants examined.

The Brazilian Caatinga is a tropical semiarid vegetation biome with a climate characterized by elevated temperatures and reduced precipitation. Frasson et al. (2012) studied the anti-trichomonas activity of 44 aqueous extracts of 23 Brazilian plants belonging to 14 different families used in folk medicine found in the Caatinga desert (Table 4).

Only the extract of roots of *Polygala decumbens* showed a significant effect on the viability of trophozoites. The extract was active against metronidazole-resistant species with MIC 1560 μg/mL.

Recently, Silva et al. (2020) studied various extracts of fruits of *Poincianella* (*Caesalpinia*) *microphylla,* a plant found in Brazilian Caatinga. The crude aqueous extract was successively extracted with different ratios of the water-methanol mixture. The main compounds identified were hydrolyzable tannins like gallotanins and ellagitannis. Procyanidin dimer, epicatechin, ellagic acid, and *O*-(digalloyl) quinic

Table 3

Various Central and South American plants and their extracts assayed for *in vitro* anti-trichomonas activity by Muelas-Serrano et al. (2000).

Plant family	Name of the plant, part of the plant used, and type of extract
Acanthaceae	*Ruellia tuberosa* L., Leaf, Aqueous
Annonaceae	*Annona reticulata* L. Leaf Aqueous
	Annona reticulala L. Seed Aqueous
	Rollinia emarginata Schlecht Aerial parts Aqueous
Araceae	*Philodendron bipinnatifidum* Schott. Aerial parts, Aqueous, Methanolic, Methanol-insol., Butanolic, and Butanol-insol.
Asteraceae	*Baccharis trimera* (Less.) DC. Leaf, Aqueous
	Mikania cordifolia (L.) Willd. Leaf, Aqueous, Methanol-insol., Methanolic, Butanolic, Butanol-insol.
	Neurolaena lobata (L.) R. Br. Leaf, Aqueous
	Tagetes lucida Cav. Aerial parts, Aqueous
	Tithonia diversifolia Hemsl. Gray. Leaf, Aqueous
	Tridax procumbens L. Aerial parts, Aqueous
Bignoniaceae	*Crescentia cujete* L. Fruit, Aqueous
	Jacaranda mimosifolia D. Don. Leaf, Aqueous
	Tecoma stans HBK Leaf, Aqueous
	Bursera simarouba (l.) Sarg. Bark, Aqueous
Caprifoliaceae	*Sambucus mexicana* Presl. ex A. DC Aerial parts, Aqueous
Euphorbiaceae	*Croton guatemalensis* Losty. Bark, Aqueous, Methanol-insol., Methanolic, Butanolic, Butanol-insol.
Hydrophyllaceae	*Wigandia caracassana* HBK Leaf, Aqueous
Leguminosae	*Cassia occidentalis* L. Leaf, Aqueous
	Erythrina crista-galli L. Leaf, and Bark, Aqueous
	Gliricidia sepium (Jacq.) Steud. Leaf, Aqueous
	Mimosa tenuiflora (Willd.) Poiret Bark, Aqueous
	Prosopis affinis Griseb. Bark, Aqueous
Lythraceae	*Heimia salicifolia* Link and Otto Leaf, Aqueous
Malpighiaceae	*Byrsonima crassifolia* (L.) HBK Bark, Aqueous
Moraceae	*Cecropia obtusifolia* Bertolini Leaf, Aqueous
	Cecropia pachystachya Mart. Leaf, Aqueous
Myrtaceae	*Psidium guajava* L. Leaf, Aqueous
	Phlebodium aureum (L.) John Smith Rhizome, Aqueous
Polypodiaceae	*Pontederiaceae Eichornia* crassipes (Martius) Leaf, Aqueous
Ramnaceae	*Scutia buxifolia* Reiss. Bark, Aqueous
Sapindaceae	*Allophylus edulis* (St. Hil.) Radlk. Leaf, Aqueous
	Chrysophyllum cainito L. Leaf, Aqueous
	Manilkara achras (Mill.) Fosberg Bark, Aqueous
Smilacaceae	*Smilax lundellii* Killip and Morton Root, Aqueous
Solanaceae	*Brunfelsia australis* Benth. Root, Aqueous
	Nicotiana glauca Graham Leaf, Aqueous
	Solanum nigrecens Mart. And Gal Leaf, Aqueous
	Solanum pylcomayense Morong. Aerial parts, Aqueous
Sterculiaceae	*Chiranthodendron pentdactilon* Larr. Fruit Aqueous, Methanol-insol., Methanolic, Butanolic, Butanol-insol.
Zingiberaceae	*Curcuma longa* L. Rhizome, Aqueous

Table 4
Plant species from the Brazilian Caatinga region and part of the plant used for screening anti-trichomonas activity by Frasson et al. (2012).

Family	Name of the plant	Part of the plant used for testing
Anacardiaceae	*Myracrodruon urundeuva* Alemão	Branch, bark, leaf
Apocynaceae	*Allamanda blanchetii* A.DC.	Branch, leaf
Burseraceae	*Commiphora leptophloeos* (Mart.) J.B. Gillett	Branch, bark
Cactaceae	*Melocactus zehntneri* (Britton and Rose) Luetzelb.	Leaf, root
Combretaceae	*Buchenavia tetraphylla* (Aubl.)	Leaf
Euphorbiaceae	*Jatropha mutabilis* (Pohl)	Branch, root
Fabaceae-Caes	*Chamaecrista desvauxii* (Collad.) Killip	Branch, fruit, leaf
	Libidibia ferrea (Mart.ex Tul.)	Fruit, leaf
	Parkinsonia aculeata L	Leaf
	Senna macranthera (Collad.)	Fruit
	Senna splendida (Vogel.)	Branch
Fabaceae-Mim	*Anadenanthera colubrina* (Vell.)	Branch, bark, fruit, leaf
	Piptadenia viridiflora (Kunth)	Branch, fruit
	Pityrocarpa moniliformis (Benth.)	Leaf
Fabaceae-Pap	*Bauhinia acuruana* Moric.	Branch, fruit, leaf
	Dioclea grandiflora Mart. ex Benth	Branch, fruit, leaf
	Myroxylon peruiferum L.f. Bálsamo	Leaf
Malpighiaceae	*Stigmaphyllon paralias* A. Juss	Leaf
Malvaceae	*Sida galheirensis* Ulbr. Malva-veludo	Branch, leaf
Myrtaceae	*Eugenia brejoensis*	Leaf
Ochnaceae	*Ouratea blanchetiana* Engl.	Branch, leaf
Polygalaceae	*Polygala boliviensis*	Branch, inflorescence, leaf
	Polygala decumbens Aubl.	Root

Table 5
IC$_{50}$ values of various plants methanolic extracts along with their families analyzed by Calzada et al. (2007).
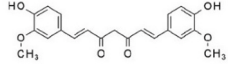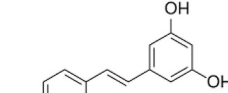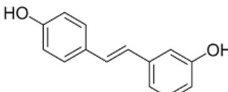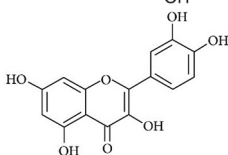
Plant family	Name of the plant	Part of the plant studied	IC$_{50}$ (μg/mL)
Anacardiaceae	*Schinus molle* L.	Aerial part	790.3
Annonaceae	*Annona cherimola* Miller	Seeds	270.9
Asteraceae	*Artemisia absinthium* L.	Aerial part	708.6
	Artemisia ludoviciana Nutt	Aerial part	230.9
	Chrysactinia mexicana A. Gray	Aerial part	105.9
	Matricaria recutita L.	Aerial part	559.9
Caricaceae	*Carica papaya* L.	Seeds	5.6
Chenopodiaceae	*Chenopodium ambrosioides* L. Green variety	Aerial part	996.7
	Chenopodium ambrosioides L. Red variety	Aerial part	105.9
	Chenopodium murale L.	Aerial part	196.1
Convolulaceae	*Dichondra argentea* Humb and Bonpl	Aerial part	317.1
Geraniaceae	*Geranium mexicanum* HB and K.	Roots	56.0
Fabaceae	*Caesalpinia pulcherrima* (L.) Sw	Aerial part	137.7
	Senna villosa Mills	Aerial part	223.9
Lamiaceae	*Thymus vulgaris* L.	Aerial part	126.4
	Ocimum basilicum L.	Aerial part	204.6
Papaveraceae	*Bocconia frutescens* L.	Aerial part	30.9
Arecaceae	*Cocos nucifera* L.	Husk fibre	5.8
Punicaceae	*Punica granatum* L.	Exocarpo of fruit	100.9
Rutaceae	*Ruta chalepensis* L.	Aerial part	226.7
Schizaeaceae	*Lygodium venustum* Sw	Aerial part	60.9
Verbenaceae	*Lippia alba* (Mill.) N.E.Br.	Aerial part	227.9
	Aloysia triphylla Britton	Aerial part	103.0

acid were also identified. Tannins enriched fractions presented anti-trichomonas activity.

Beas of *Phaseolus vulgaris* L., (Perla black bean) a staple food in many Latin American countries, are largely consumed because of their high protein content. IC$_{50}$ of acidified water and acetic acid extract of perla black bean for *in-vitro* antitrichomonal activity against *T. vaginalis was* 176.8 and 378.3 μg/mL, respectively, showing the potential of this plant (Lara-Díaz et al., 2009). *Arbutus unedo*, a European plant, is now naturally adapted to dry summer climates and is now cultivated as an ornamental plant in California and North America's west coast. Ethyl acetate extract of *Arbutus unedo* leaves was an effective anti-trichomonacidal with 100% growth inhibition of *T. vaginalis* at 500 μg/mL (Ertabaklar et al., 2009).

3.3.3. Mexican plants

In a search for new chemotherapeutic agents for trichomoniasis, Calzada et al. (2007) screened crude methanolic extracts of 22 Mexican medicinal plants commonly used for abdominal pain, colic, and vaginal discharge for antitrichomonal activity against *T. vaginalis*. In all, 22 plants belonging to 15 different families were assayed. The details of the plants and important experimental findings are shown in Table 5.
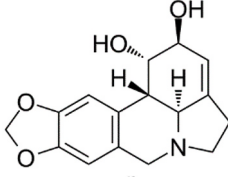
As observed in Table 5, *Carica papaya* and *Cocos nucifera*, showed the best anti-trichomonas activity with IC$_{50}$ values of 5.6 and 5.8 μg/mL, respectively. Moderate activity was observed with the species *Bocconia frutescens*, *Geranium mexicanum*, and *Lygodium venustum* with IC$_{50}$ values 30.9–60.9 μg/mL. The rest of the plants are considered ineffective as the IC$_{50}$ values were considerably greater than 100 μg/mL. Although effective anti-trichomonal, all plants were still very less effective than metronidazole with 0.037 μg/mL.

Extract of peels of pomegranate fruits; *Punica granatum* were also tested by El-Sherbiny and El Sherbiny (2011) for *in-vitro* anti-*T. vaginalis* activity. In this study, the authors also determined the *in-vivo* anti-trichomonal activity of *Commiphora molmol* (Engl.) Engl. ex Tschirch (Mirazid) in metronidazole and tinidazole resistant females, and the dose was two capsules (600 mg) for six to eight successive days on an empty stomach. Both these plants showed promising effects.

3.3.4. African plants

On similar lines, scientists from South Africa (van Vuuren and Naidoo, 2010) investigated several plants indigenous or introduced to southern Africa and are used by ethnic rural populations to treat infections related to the urogenital tract *Aloe ferox* Mill. leaves, *Bowiea volubilis* bulb, *Bowiea volubilis* Harv. leaves *Carpobrotus edulis* (L.) L. Bolus leaves *Cassia occidentalis* Náves leaves *Cassia occidentalis* Náves seeds *Catharanthus roseus* (L.) G. Don leaves, *Cissus quadrangulis* leaves, *Croton gratissimus* Burch. leaves, *Euclea natalensis* A. DC. leaves, *Hypericum aethiopicum* Thunb. roots, *Hypericum aethiopicum* Thunb leaves *Polygala fruticosa* P.J. Bergius leaves, *Psidium guajava* L. leaves, *Sansevieria aethiopica* Thunb. leaves, *Scabiosa columbaria* L. roots, leaves, *Strelitzia reginae* Aiton leaves, *Syzygium cordatum* Hochst., *Tarchonanthus camphoratus* L. leaves, *Terminalia sericea* Burch. ex DC. leaves, *Typha capensis* (Rohrb.) NE Br. leaves, Essential oil of *Croton gratissimus* Burch., and *Tarchonanthus camphoratus* L. Aqueous extracts of these plants were not as effective as observed from their high MIC values. In organic extracts prepared using dichloromethane and methanol (1:1), only three plants' meaningful activity leaves extract of *Psidium guajava* L., *Strelitzia reginae* Aiton, and *Tarchonanthus camphoratus* L. Later, in 2013, the same group (Naidoo et al., 2013) extensively studied several plants used individually or in combination for the treatment of sexually transmitted diseases in rural northern Maputaland, KwaZulu-Natal was explored. Plants were selected based on their applications and use in traditional medicine. Aqueous and organic extract of the following plants were studied for cytotoxicity and antimicrobial activity against various pathogens including *T. vaginalis*. *Albizia adianthifolia* (Schumach.) W. Wight, *Aloe marlothii* Berger, *Bidens pilosa* L., *Carica papaya* L., *Clematis brachiate*, *Euphorbia hypericifolia* Boiss., *Hypoxis hemerocallidea* Fisch. and C.A. Mey., *Kigelia Africana* (Lam.) Benth., *Musa acuminate*, *Ozoroa engleri* R. Fern. and A. Fern., *Peltophorum africanum* Sond., *Ranunculus multifidus* Forssk., *Sarcophyte sanguinea* Sparrm., *Sclerocarya birrea* (A. Rich.) Hochst., *Senecio serratuloides* DC., *Syzygium cordatum* Hochst, *Tabernaemontana elegans* Stapf, *Trichilia dregeana* Sond., and *Ximenia caffra* Sond. In this study, aqueous extracts were ineffective, as seen from
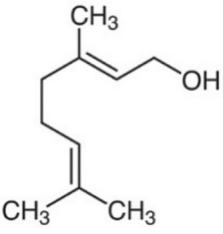
Table 6
An overview of several plants and phytochemicals with promising anti-trichomonas activity.

Name	Origen	Source	Chemical structure	Effect	Remarks
Polyphenol Curcumin	polyphenol extracted from the Indian spice turmeric			possessed anti-protozoal activity two reports are suggesting a contradictory effect of curcumin on *T. vaginalis*	curcumin is a well-tolerated compound and is safe in at least six human clinical trials
Polyphenol Resveratrol	dietary non-flavonoid polyphenolic phytoalexin	skin of grapes		inhibited the growth of *T. vaginalis* cytostatic effect, whereas higher doses exerted killing effects	Deleterious effects as anti-trichomonal via hydrogenosomal dysfunction and metabolic alteration
Anthraquinone Emodin	a derived anthraquinone, *cuspidatum*	is extracted from roots and barks of various plants like *Rheum palmatum, Aloe vera, Polygonum multiflorum,* and *Polygonum*		emodin significantly decreased the number of trichomonads in the vagina as well	cured the trichomonal abscesses, indicating good absorption of emodin anthraquinone.
Flavonoid Quercetin Quercetrin	Extract of leaves and twigs of both the *Dorstenia* species. Extracts of flowers, leaves, and stem of *Rheum ribes* L.	*Dorstenia barteri* and *Dorstenia convexa* extracts and *Rheum ribes* L.		Quercetin was found to be most potent, followed by quercitrin	anti-trichomonal activity was attributed to flavonoids, namely quercetin, quercitrin
Alkaloid lycorine	Plants of Amaryllidaceae family			arrested the *T. vaginalis* cell cycle at the G2/M phase of the cell cycle	
Alkaloid Candimine	bulbs of *Hippeastrum morelianum*			modifications in size, shape, and intracellular localization of hydrogenosomes	arrest of the *T. vaginalis* cell cycle; hydrogenosome autophagy
Alkaloid lycorine-Candimine				*T. vaginalis* NTPDase and ecto-5′-nucleotidase activities were significantly inhibited by candimine and lycorine	
Monoterpenoid geraniol	oil of *Amomum tsao-ko*	a spice used in Chinese cuisine and is also part of the Chinese Traditional Medicine		10-fold less active than metronidazole	Cell and organelles damage, Intense morphological changes

Table 6 (*continued*)

Name	Origen	Source	Chemical structure	Effect	Remarks
			CH$_3$ — OH — CH$_3$ CH$_3$ (structure)		
β glycosides	Thai plants	Seeds, flowers, leaves, fruits		125-fold less effective than metronidazole	potent anti-trichomonal with MIC of 6.25 μM.
Diterpenoids and triterpenoids	barks of *Platanus acerifolia* and *Malus domestica*	peels	Betulinic acid and ursolic acid	betulinic acid presented better activity than those from ursolic acid	betulinic acid: better activity than ursolic acid. Piperazine derivatized from betulinic acid: high trichomonacidal agents.
Saponins	*Passiflora alata; Quillaja brasiliensis; Ilex paraguariensis; Sapindus mukorossi*	Leaves; fruit pericarp of *Sapindus mukorossi*		MIC for saponins was four-fold	The combination of saponin with synthetic spermicide resulted in lowering the anti-trichomonal MIC
Essential oils	*Lavandula angustifolia* and *Lavandula intermedia*		*M. piperita* and *S. officinalis*: anti-trichomonas activity	Comparable to metronidazole *S. officinalis* was found to have higher potency as compared to *M. piperita*	*Lavandula angustifolia* and *Lavandula intermedia* were evaluated for their anti-trichomonal properties
Eucalyptus	*Eucalyptus camaldulensis*	aqueous extract of *E. camaldulensis* leaves		eradication of *T. vaginalis* at the end of 24 h	vaginal cream containing extract of *E. camaldulensis* along with extract of *Viola odorata* and *Mentha piperita*
American plants	*Mikania cordifolia Neurolaena lobata*	Asteraceae and *Scutia buxifolia* Reiss from the Ramnaceae		The most active extracts amongst all the plants examined	
Brazilian Caatinga region	*Polygala decumbens* 0 μg/mL.	roots		Significant effect on the viability of trophozoites.	The extract was active against metronidazole-resistant species with MIC 156
California	*Arbutus unedo*	ornamental plant		effective anti-trichomonacidal	100% growth inhibition of *T. vaginalis* at 500 μg/mL
Mexican plants	*Carica papaya* and *Cocos nucifera*,			anti-trichomonas effect	less effective than metronidazole
African plants	*Cussonia* species (Araliaceae)	Methanolic extract of leaves		Anti-trichomonas activity against *T. vaginalis*	MIC in the range of 0.8–1.3 μg/mL

their high MIC values (>16 mg/mL) whereas organic extracts exhibited MIC from 2 to 12 mg/mL.

Cussonia species (Araliaceae) are used in African traditional medicine to treat pain, inflammation, gastro-intestinal disorders, malaria, and sexually transmitted diseases. Methanolic extract of leaves of 13 *Cussonia* species was tested for *in vitro* anti-trichomonas activity against *T. vaginalis,* and MIC was found to be in the range of 0.8–1.3 μg/mL (De Villiers et al., 2010).

4. Concluding remarks

Trichomoniasis is considered a re-emerging disease, and drug resistance against metronidazole or 5-nitro imidazole family, the first-line drug for treating trichomoniasis, is on the rise. Lack of alternative treatment options for trichomoniasis is a serious clinical threat, and there is a critical need to have alternative drugs for its management. Traditional/folk medicines that are in practice in various countries have carved out the pathway for anti-trichomonal drug research. Whole-plant extracts and phytoconstituents form a major part of these traditional anti-trichomonal medicines. Negligible adverse events are one of the major benefits of using plant-based medicines. Much research has been carried out on such phytoconstituents or extracts, and their therapeutic utility has been established. However, most of this research is sparse and is mostly limited to *in-vitro* studies. These studies have revealed several promising phytoconstituents or plant extracts that need to be evaluated in experimental models and clinical settings. Many potential phytochemicals have been evaluated for them *in-vitro* anti-trichomonas activity but have not been explored subsequently. It is desired that further research should focus on the clinical efficacy of these phyto-constituents or plant extracts. Concerted efforts are needed to take these potential phytoconstituents/plants for therapeutic usage.

Our overview indicates that several plants and phytochemicals have promising anti-trichomonas activity (Table 6).

However, considering the limited quality of included evidence and heterogeneity of different studies, more well-design studies are required to further confirm the conclusion. We recommend that future studies focus on the intensity and treatment duration of selected plant material or isolated phytoconstituents and pay more attention to safety profiles. Moreover, a better understanding of the selected active constituents' mechanism of action is needed to help researchers and clinical professionals pursue further studies. Structure-activity relationship studies will also help identify the essential structural features of the selected molecule.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper. All authors have seen and approved the final version of the manuscript and warrant that the article hasn't received prior publication and isn't under consideration for publication elsewhere.

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