JSIANE: Vol. 4, Issue 4, 2024, Page 75-80 ISSN (Online) 2583-2093

Journal of Science Innovations and Nature of Earth

Journal homepage : www.jsiane.com

Qualitative Analysis of Crude Extract from the Leaves of *Cassia occidentalis*

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DOI: https://doi.org/10.59436/jsiane.196.2583-2093

Abstract

Medicinal plants have intrinsic antioxidant qualities that these items use to enhance health, prevent sickness, and prolonged life expectancy. In this study, a preliminary study of qualitative analysis was performed in crude extracts of leaf extracts of *Cassia occidentalis* using various solvent on the basis of polarity. The data obtained in the *in vitro* models clearly establish the antioxidant potency of leaf extracts of *Cassia occidentalis*. In the present study we have used aqueous extract of the leaves, followed by the methanolic, disteled water and petroleum ether extracts, respectively. We have reported the presence of carbohydrates, alkaloids, phenols, tannins, phytosterols, and flavonoids through a series of standard tests. The study provides substantial evidence supporting the therapeutic potential of *Cassia* extract, as sources of various natural antioxidants and anticancer agents. The exploration of these plant extract could potentially lead to the development of new, effective, and natural therapeutic agents for the treatment of various diseases, particularly those related to oxidative stress and cancer.

Keywords: *Cassia occidentalis*, Crude Extract, Qualitative Analysis, Antioxidants, Anticancer agents, Medicinal plants.

Received 25.09.2024 **Revised 26.10.2024** **Accepted 30.12.2024**

Introduction

Traditional medicine has been used for the treatment of human illnesses since ancient times and is mainly based on components derived from natural products, from herbs, plants and animals. A number of traditional systems of medicine exist in India, which include Ayurveda, Siddha, and Unani. In India, the concept of the Ayurvedic system of medicine was developed between 2500 and 500 BC. Traditional medicine system has been mentioned in the ancient Vedas and other scriptures (Pandey *et al.*, 2013, Valsaraj *et al.*, 1997). Several previous experiments have shown that antioxidant supplements are useful in cancer treatment (Valko *et al.*, 2007). Polyphenols, flavonoids, and brassinosteroids are examples of plant secondary metabolites that have been the subject of much research due to their potential anticancer effects. By combining their antioxidant, anti-proliferation, anti-cancer, anti-cell- selectivity, and anti-cell-killing effects, these compounds have shown anticancer action (Cao *et al.*, 2013; Gupta *et al.*, 2014). Traditional healers have long recognised the therapeutic potential of plants for a variety of medical conditions. Reduce inflammation, kill germs, and fight cancer—just a few of the many recognize d medicinal benefits offered by these herbs. Cardiovascular disease and other severe ailments have also been treated with them. As resistance to cancer treatments has increased, researchers have increasingly turned to natural chemicals derived from plants and the ocean. Plant extract acts synergistically (Kumar and Arya, 2022). It is now generally accepted that plants' positive benefits are not due to a single chemical but rather to a complex interaction between the composite combination of molecules contained in the whole plant, even though several substances extracted from plants have been tested for anticancer activities. *Cassia occidentalis* is often known in Hindi as Kasunda or Bari kasunda; it is a member of the Leguminosae family (Yadav, 2011). The antioxidant properties of medicinal plants have long been praised as a crucial defence against oxidative stress, a key component in several diseases (including cancer). Cells are damaged by oxidative stress, which occurs when the amount of reactive oxygen species (ROS) in the body is too high compared to its ability to neutralise them. Medicinal herbs are rich in antioxidants, which destroy harmful ROS and reduce the likelihood of chronic diseases and cell damage (Marinescu *et al.*, 2014). Since their protective capabilities have been well recognised, research into these antioxidants has mostly concentrated on dietary supplements, cosmetics, and pharmaceuticals. Antioxidants are essential for warding off oxidative stress, which is linked to several degenerative diseases and the natural ageing process. Deficiency in antioxidant defences causes reactive oxygen species (ROS) levels to rise, leading to oxidative stress. This dysregulation harms cells, exacerbating conditions including cardiovascular disease, cancer, and

neurological disorders. Antioxidants protect cells and tissues from more oxidative damage by eliminating reactive oxygen species (ROS). Plants are a rich source of phytochemicals and act as a larvicide against mosquitoes (Kumar and Arya, 2022), Alkali et *al.*(2024) concluded that the extracts of *Cassia occidentalis* are a rich source of phytochemicals. The most prevalent phytochemical is found to be flavonoids, while alkaloids are also present in very high concentrations. The phytochemical screening revealed the presence of bioactive constituents that could be the reason for the pharmacological activity that is used traditionally by many people as an alternative treatment for a variety of health diseases. Haifa (2022) studied in *Cassia* and *Senna* species could be a promising source of natural bioactive agents with beneficial effects for human health. Ntchapda (2015) suggest that *Cassia occidentalis* aqueous extract has diuretic and antioxidant activities, and deserves further studies considering the potential for the treatment of hypertension. Keeping in view of above properties, we have designed this study to analyze phytochemical constitutents in the leaves extract of *Cassia occidentalis* using various extraction solvents.

Material and Methods

The identification process confirmed that the plants were indeed Cassia occidentalis consistent with their known botanical characteristics. In this study, the medicinal plants Cassia occidentalis were meticulously collected from various ecological niches across the Ajmer District, located in the state of Rajasthan, India. Following identification, the plants were authenticated, and voucher specimens were prepared. These **specimens were pressed, dried, and mounted on herbarium sheets,** which were then labeled with detailed information, including the plant's scientific name, collection site, date of collection, and the name of the collector.

Preparation of Extracts:- The extraction of phytochemicals from *Cassia occidentalis* leaves was performed through a methodical process designed to maximize the yield of bioactive compounds while preserving their chemical integrity.The dried and powdered plant material was divided into three portions, each designated for extraction with one of the selected solvents: water, methanol, and petroleum ether The use of different solvents allows for the selective extraction of compounds based on their polarity (Pandey and Tripathi, 2014; Sasidharan *et al.*, 2011):-

Water (Polar): Used for extracting hydrophilic compounds such as alkaloids, glycosides, tannins, and certain flavonoids. Water is particularly effective in extracting compounds that are highly soluble in aqueous environments, including many antioxidants and polysaccharides.

Methanol (Mid-polar): Methanol is an effective solvent for extracting a broad range of bioactive compounds, including both polar and nonpolar substances. It is particularly useful for isolating phenolic compounds, flavonoids, and certain saponins, which have intermediate polarity.

Petroleum Ether (Non-polar): Petroleum ether is used to extract nonpolar compounds, such as lipids, fatty acids, terpenoids, and some alkaloids. This solvent target hydrophobic compounds that are not soluble in water or methanol (Pandey and Tripathi, 2014).

Extraction Process:

Weighing and Mixing: Approximately 50 gms of the powdered plant material was accurately weighed for each solvent extraction. The plant powder was then mixed with 500 mL of the respective solvent in a clean, dry conical flask. The ratio of plant material to solvent was carefully chosen to optimize the extraction efficiency while ensuring that the solvent could fully penetrate the plant matrix.

Maceration: The mixtures were subjected to maceration, where they were allowed to stand for 48-72 hours at room temperature with intermittent shaking. Maceration is a passive extraction technique that allows the solvent to permeate the plant cells, dissolving the phytochemicals within. The flasks were covered with aluminum foil to prevent contamination and solvent evaporation. The prolonged contact time between the plant material and solvent ensures maximum extraction of bioactive compounds.

Filtration: After the maceration period, the mixtures were filtered to separate the liquid extract from the solid plant residue. Filtration was performed using Whatman No. 1 filter paper and a Buchner funnel. The filtration process was conducted slowly to ensure that all the extract was collected and that the filter paper did not clog, which could lead to loss of material.

Concentration of Extracts: The filtered extracts were then concentrated using a rotary evaporator under reduced pressure at a temperature not exceeding 40°C. This process removes the bulk of the solvent, leaving behind a concentrated extract that is rich in the bioactive compounds. The low-temperature evaporation minimizes the risk of thermal degradation of sensitive phytochemicals.

Drying of Extracts: After concentration, the extracts were transferred to pre-weighed glass dishes and further dried in a vacuum oven to remove any remaining traces of solvent. The drying process was continued until constant weight was achieved, ensuring that the extracts were completely free of solvent.

Storage of Extracts: The dried extracts were collected, weighed, and stored in airtight amber glass vials to protect them from light and moisture, which could lead to degradation of the active compounds. The vials were labeled and stored at 4°C until further use in various assays and analyses. The methods employed in this study are consistent with best practices in phytochemical research, ensuring the reliability and reproducibility of the results (Abubakar and Haque (2020).

Qualitative Analysis Parameters:-

Qualitative tests for primary and secondary metabolites are essential for the preliminary screening of phytochemicals present in plant extracts. These metabolites include carbohydrates, phenols, flavonoids, alkaloids, tannins, and phytosterols, all of which contribute to the medicinal properties of plants. The following section provides an in-depth overview of the qualitative tests used in this study, along with their procedures and expected results.

Result and Discussion

Extraction- In this study, the extraction of bioactive compounds from *Cassia occidentalis (CO)* was conducted using different solvents to target a broad spectrum of polarities. Water (polar), methanol (mid-polar), and petroleum ether (non-polar) were employed as solvents in a cold extraction method. This technique ensures that sensitive compounds are not degraded by heat, maintaining their

bioactivity. The yield and physical properties of the extracts varied significantly based on the solvent used, reflecting the diverse nature of the compounds being targeted.

(1)

Table: Physical Properties of the Obtained Extract

S.N.	Sample Name	Extract Weight (mg/g.dw)	Extract Colour	Appearance
1	CO (M)	381	Dark Green	Sticky
2	CO (W)	591	Lightish Brown	Powder
3	CO (PE)	297	Dark Brown	Sticky

The physical properties of the extracts obtained from *Cassia occidentalis* (CO) using different solvents—Methanol (M), Water (W), and Petroleum Ether (PE)-were meticulously documented to provide insights into their consistency and potential applications. The extractive values, which indicate the quantity of extract obtained per milligram per gram of dry weight of plant material, varied significantly across the different solvent systems, reflecting the differential solubility of the phytochemicals in these solvents. For the methanolic extracts, *Cassia occidentalis* (CO) yielded a substantial amount of 381 mg/g.dw, characterized by a dark green color and a sticky appearance. This high yield suggests that methanol effectively extracted a considerable amount of bioactive compounds from the *Cassia* plant material, which could include various polyphenols, alkaloids. The water extracts, which are expected to solubilize more polar compounds, yielded different results. *Cassia's* water extract (CO (W)) had the highest extractive value among all samples, at 591 mg/g.dw, and was characterized by a lightish brown color and a powdery appearance. The significant yield and distinct color suggest a rich composition of water-soluble compounds, such as tannins, glycosides, and polysaccharides, which are commonly extracted by aqueous solvents. This lighter color compared to *Cassia's* water extract might reflect a different profile of water-soluble compounds, potentially with less polyphenolic content or different types of flavonoids. Petroleum ether, a non-polar solvent, was used to extract more lipophilic components. *Cassia's* petroleum ether extract (CO (PE)) resulted in 297 mg/g.dw and was dark brown and sticky, indicating the extraction of non-polar compounds such as lipids, terpenes, and possibly waxes. These compounds often impart a darker color and a sticky texture due to their hydrophobic nature. The yellow hue may suggest the presence of certain terpenoids or carotenoids, which are typically soluble in non-polar solvents. The variation in extractive yields, colors, and textures across the different solvents underscores the diversity of phytochemicals present in *Cassia*. These physical characteristics not only provide initial clues about the chemical nature of the extracts but also help in determining the suitability of each extract for further chemical analysis and potential therapeutic applications. The sticky versus powdery nature of the extracts could also influence their handling and formulation in practical applications, such as in the preparation of herbal medicines or supplements.

Qualitative Test of Extracts- The qualitative analysis of *Cassia occidentalis* extracts across various solvents provided a comprehensive view of the phytochemical composition. The presence of carbohydrates, alkaloids, phenols, tannins, phytosterols, and flavonoids was confirmed through a series of standard tests, each tailored to detect specific functional groups or compound classes.

Plate 4.1: Plant material in different solvents.

Plate 4.2: Filtered Extracts.

Plate 4.3: Dry Extracts

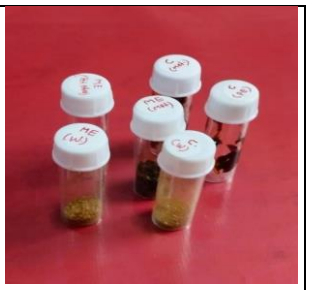


Plate 4.4: The Collected

All methanolic and water extracts of both plants tested positive across all phytochemical tests, indicating a rich presence of these bioactive compounds. Specifically, Fehling's test for carbohydrates, Mayer's test for alkaloids, $FeCl_3$ test for phenols, and gelatin test for tannins all yielded positive results, underscoring the plants' "potential therapeutic properties. These findings are consistent with the traditional uses of these plants in herbal medicine, where such compounds are known to exhibit a wide range of bioactivities, including antioxidant, anti- inflammatory and antimicrobial effects".

Interestingly, the petroleum ether extracts showed a selective presence of phytochemicals. In both *Cassia occidentalis*, the petroleum ether extracts tested negative for carbohydrates, alkaloids, and phenols but positive for tannins and phytosterols. This selectivity is typical of non-polar solvents, which are more efficient in extracting lipophilic compounds like phytosterols and certain tannins. These results provide a basis for focusing further analyses on specific extracts,

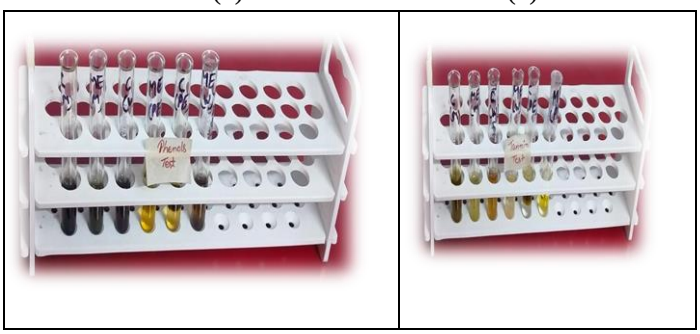
depending on the desired bioactivity. For instance, the methanolic extracts, rich in a wide range of phytochemicals, Bagega (2018) studied *Cassia occidentalis* (CO) Leaf Extract on antibacterial activity and antioxidant activity against *Salmonella typhimurium*. Kumar and Arya (2022) examine the synergistic larvicidal action of leaf and seeds of different plant extracts as a larvicide against mosquitoes. In this plant extract some compounds are present in a trace amount, whereas saponin was present in high amount. In addition to these flavonoids were found in a moderate amount, while tannins, glycoside, cardiac glycosides, steroids, saponin glycoside, anthraquinones and volatile oil were present in trace amount.

Table: Qualitative Test Results of Extracts

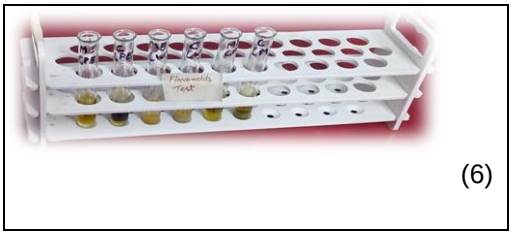
Sample name	**Carbohydrate test**	**Alkaloids test**	**Phenols test**	**Tannin test**	**Phytosterol test**	**Flavonoids test**
Test name	**Fehling's test**	**Mayer's test**	**$FeCl_3$ test**	**Gelatine test**		**Lead Acetate test**
CO (M)	Positive	Positive	Positive	Positive	Positive	Positive
CO (W)	Positive	Positive	Positive	Positive	Positive	Positive
CO (PE)	Negative	Negative	Negative	Positive	Positive	Negative

The results demonstrated a comprehensive presence of these bioactive compounds in the methanolic (M) and water (W) extracts of plant. Specifically, all tests conducted on the methanolic and aqueous extracts of *Cassia occidentalis* (CO (M) and CO (W)) returned positive results. This indicates a rich phytochemical composition, with extracts containing significant amounts of carbohydrates, alkaloids, phenols, tannins, phytosterols, and flavonoids. Similarly, the phytochemical profile of Tinospora cordifolia has shown the different phytoconstituents in different solvent extracts (Kumar and Arya, 2023). The consistent positivity across these tests suggests that methanol and water are effective solvents for extracting a wide range of bioactive compounds from these plants. Methanol, being a mid-polar solvent, is known for its ability to extract a broad spectrum of compounds, including polar and some non-polar substances, which is reflected in the comprehensive extraction of secondary metabolites like alkaloids, phenols, and flavonoids. The water extracts similarly showed robust positive results, particularly for water-soluble compounds such as carbohydrates and tannins, which are known for their solubility in polar solvents like water. In contrast, the petroleum ether (PE) extracts of *Cassia* plants exhibited a more selective profile. The *Cassia occidentalis* petroleum ether extract (CO (PE)) tested negative for carbohydrates, alkaloids, and phenols, indicating that these non-polar solvents were less effective at extracting these polar compounds. However, both extracts tested positive for tannins and phytosterols, which are less polar than other compounds like carbohydrates or alkaloids, suggesting that petroleum ether was effective in extracting these particular classes of non-polar phytochemicals. The presence of phytosterols in the petroleum ether extract aligns with the expectation that non-polar solvents would extract compounds like sterols, which have a lipophilic nature. The absence of flavonoids in the petroleum ether extracts further supports the idea that these are more efficiently extracted by polar or mid-polar solvents like water and methanol, respectively. The qualitative analysis of these extracts highlights the versatility of methanol and water in extracting a wide array of bioactive compounds from *Cassia occidentalis*, while petroleum ether selectively extracts non-polar substances. This comprehensive profile of extracted phytochemicals is crucial for further studies, including quantitative analyses and bioactivity assays, to understand the full potential of these plants in pharmacological applications. The presence or absence of these compounds in different solvent extracts also provides valuable information for optimizing extraction methods based on the desired bioactive components, which is essential for the development of plant-based therapeutics. The positive results for a wide range of tests in methanol and water extracts underscore their potential as rich sources of therapeutic agents, whereas the more selective extraction by petroleum ether points to its utility in isolating specific non-polar compounds like phytosterols and tannins.

(2) (3)



(4) (5)



(6)

Plates (1-6) : Qualitative test for different Phytochemicals.

Conclusions

The results of this study underscore the significant potential of *Cassia occidentalis* (CO) extract as sources of bioactive compounds with various therapeutic properties.

The extraction process, conducted using solvents of varying polarities (water, methanol, and petroleum ether), provided a diverse array of extracts with distinct physical properties and chemical compositions. The differences in extract yields and characteristics-such as color, texture, and weight—highlight the efficacy of different solvents in extracting specific types of compounds. For instance, methanol, a mid-polar solvent, was particularly effective in extracting a substantial amount of bioactive compounds from *Cassia*, as indicated by the high extractive value and the sticky, dark green appearance of the extract. The qualitative analysis of the extracts provided further insights into their phytochemical compositions, confirming the presence of key bioactive compounds such as carbohydrates, alkaloids, phenols, tannins, phytosterols, and flavonoids. The methanolic and aqueous extracts of both plants showed a rich presence of these compounds, with all tests returning positive results. This comprehensive presence suggests that *Cassia* have a wide range of therapeutic potential, as these compounds are known for their antioxidant, anti- inflammatory and antimicrobial properties. The petroleum ether extracts, however, displayed a more selective phytochemical profile, highlighting the effectiveness of non-polar solvents in extracting specific compounds like phytosterols and certain tannins, which are associated with cholesterol-lowering and anti-inflammatory effects. Abd Elkarim, (2024) has also investigated that Petroleum Ether Extract of *purslane* leaf has anti-inflammatory, antioxidant and anticancer activities. The study provides substantial evidence supporting the therapeutic potential of *Cassia occidentalis* leaves extract, as sources of natural antioxidants and anticancer agents. The exploration of these plant extracts could potentially lead to the development of new, effective, and natural therapeutic agents for the treatment of various diseases, particularly those related to oxidative stress and cancer.

References

Abd Elkarim, A.S., Mohamed, S.H., Ali, N.A., Elsayed, G.H., Aly M.S., Elgamal, A.M., Elsayed, W.M. and El-Newary, S.A. (2024). The Phytochemical Profile of the Petroleum Ether Extract of *Purslane* Leaves and Its Anticancer Effect on 4-(Methylnitrosamino)-1-(3-pyridyl)-1-buta-4 None (NNK)-Induced Lung Cancer in Rats. Int J Mol Sci. Dec 4; 25(23), 13024.

Abubakar, A. R., and Haque, M. (2020). Preparation of Medicinal Plants: Basic Extraction and Fractionation Procedures for Experimental Purposes. Journal of Pharmacy & Bioallied sciences, *12*(1), 1–10.

Alkali K., Dikwa K. B., Ajibade, G.A., Magaji, Y., and M.B., Abdulhamid. (2024). Qualitative and Quantitative Phytochemicals Screening of Aqueous, Methanol and Hexane Leaves Extracts of *Senna Occidentalis*. Asian Plant Research Journal, Volume 12, Issue 3, Page 27-35.

Amponsah Isaac Kingsley, Mensah Abraham Yeboah, Ampofo Elikplim Kwesi, Bekoe Samuel Oppong, Sarpong Francis Mainoo and Jibira Yakubu (2016). Pharmacognostic studies of the leaves and seeds of *Cassia occidentalis* (Linn.) (Leguminosae), Journal of Pharmacognosy and Phytochemistry ; 5(3): 250-255.

Kumar, A and Arya, H (2022). Synergistic Larvicidal Action of Indigenous Plant Extracts in Combination Against *Aedes aegypti* (Diptera: Culicidae), Journal of Science Innovations and Nature of Earth; 2(2): 17-22.

Kumar, A and Arya, H (2022). Phytochemical analysis and synergistic larvicidal action of Argemone mexicana against third instar larvae of *Aedes aegypti* (Diptera: Culicidae), Journal of Science Innovations and Nature of Earth; 2(1): 14-20.

Kumar, A and Arya, H (2023). Phytochemical screening and larvicidal evaluation of leaf extract of Tinospora cordifolia against *Aedes aegypti* (Diptera: Culicidae), International Journal of Entomology Research: 8(4): 11-19.

Bagega A.I., Usman A.A., Dankaka S.M., Amina M. and Mani A.U.1 (2018). Antibacterial Activity and Phytochemical Analysis of *Cassia occidentalis* Leaf Extract on *Salmonella typhimurium*, Bioremediation Science And Technology Research, Vol. 6, Page No 2, 5-8

Cao, G., Sofic, E., & Prior, R. L. (2013). Antioxidant capacity of tea and common vegetables. Journal of Agricultural and Food Chemistry, 44(11), 3426-3431.

Gupta, S. C., Tyagi, A. K., Deshmukh-Taskar, P., Hinojosa, M., Prasad, S., & Agarwal, B. B. (2014). Downregulation of tumor necrosis factor and other pro-inflammatory biomarkers by polyphenols. Archives of Biochemistry and Biophysics, 559, 91-99.

Haifa A A., Omer, Giovanni Caprioli, Doaa Abouelenein, Ahmed M Mustafa, Abdullahi Ibrahim Uba, Gunes Ak, Refiye Beyza Ozturk, Gokhan Zengin, Sakina Yagi (2022). Phenolic Profile, Antioxidant and Enzyme Inhibitory Activities of Leaves from Two *Cassia* and Two *Senna* Species., Molecules. 2022 Aug 30;27(17):5590.

Marinescu, B., Dehelean, C. A., Rusu, L. C., Gheorgheosu, D., Muntean, D., & Cinta-Pinzaru, S. (2014). A comparative study: St. John's Wort (Hypericum perforatum) and its active compounds in experimental colon cancer. Pharmaceutical Biology, 52(6), 723-730.

Ntchapda Fidèle , Barama Joseph , Azambou David Romain Kemeta, Etet Paul Faustin Seke , Dimo Théophile (2015). Diuretic and antioxidant activities of the aqueous extract of leaves of *Cassia occidentalis* (Linn.) in rats. Asian Pacific Journal of Tropical Medicine.Volume 8 (9), September, 685-693.

Pandey, M. M., Rastogi, S., & Rawat, A. K. S. (2013). Indian traditional Ayurvedic system of medicine and nutritional supplementation. Evidence-Based Complementary and Alternative Medicine, 2013, Article ID 376327.

Pandey, A and Tripathi, S. (2014). Concept of standardization, extraction, and pre-phytochemical screening strategies for herbal drug. Journal of Pharmacognosy and Phytochemistry, 2, 115–119.

Valko, M., Leibfritz, D., Moncol, J., Cronin, M. T., Mazur, M., & Telser, J. (2007). Free radicals and antioxidants

in normal physiological functions and human disease. The International Journal of Biochemistry & Cell Biology, 39(1), 44-84.

Valsaraj, R., Pusphangadan, P, Smitt, UW, Adsensen, A., and Nyman, U (1997): Antimicrobial screening screening of selected medicinal plants from India. Journal of Ethnopharmacology, Volume 58 (2), October, 75-83.

Yadav, R. (2011). *Cassia occidentalis*: A review on its ethnobotany, phytochemical and pharmacological profile. Journal of Pharmacy Research, 4(7), 2234-2240.

Sasidharan, S., Chen, Y., Saravanan, D., Sundram, K.M. and Yoga Latha L. (2011). Extraction, isolation and characterization of bioactive compounds from plants' extracts. Afr J Tradit Complement Altern Med. 8(1), 1-10.

Zygmunt, J.B. and Namiesnik J. (2003). Preparation of samples of plant material for chromatographic analysis. J Chromatogr Sci, 41, 109–116.