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Original Research Article **Biotechnology**

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MANGIFERA INDICA KERNEL PHYTOCHEMICALS, A POTENT REMEDY FOR ORAL-DENTAL INFECTIONS

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ABSTRACT

This study evaluates the anti-bacterial potential of *M. indica* twig and kernel phytochemicals against the common oral bacterial pathogens including *S. aureus, St. oralis, St. mutans, St. gordonii, L. brevis* and *L. casei*. The twig and kernel dried powder samples were fractionated with Soxhlet extractor with eluotrophic series of solvents viz. petroleum-ether, chloroform, absolute-ethanol, 80%-methanol and distilled-water. Antibacterial activity of the fractionated extracts was determined by zone of inhibition, minimum inhibitory concentration (MIC), minimum bactericidal concentration (MBC) and 50% inhibitory concentration (IC_{50}) values against oral pathogens while antioxidant activity was measured by $ABTS^{+}$ and DPPH assays. The results showed exceptionally high antibacterial activity in kernel ethanolic extracts (MiKE) against all the tested oral bacterial pathogens as confirmed by its highest zone of inhibition and lowest MIC, MBC and IC50 values. *M.indica* kernel extracts (MiKE) was extremely effective against *S. aureus* followed by *St. oralis, St. gordonii, L. brevis, St. mutans,* and *L. casei* with the IC_{50} value of 0.015, 0.027, 0.056, 0.081, 0.108±0.003 and 0.122±0.001 mg/ml respectively. MiKE showed excellent $ABTS^{+}$ and DPPH free radical scavenging activities with IC_{50} values of 0.649±0.0 and 0.264±0.001 μg/ml respectively amid abundance of phenolics, flavanoids and tannins. Regression analysis of antibacterial activities of fractionated extracts with phytochemical contents and antioxidant activities suggested existence of multiple pathways of inhibition in bacteria. Present study successfully establishes *M. indica* kernel extracts as an incredible and safe prospective natural remedy for oral-dental infection.

KEYWORDS: ***Antibacterial activity; antioxidant; M.indica kernel; oral bacterial pathogens; polyphenols***

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DENDROBENTHAMIDIA CAPITATA (WALL.) HUTCH. FRUIT (BHOMORA): AN OPULENT SOURCE OF POLYPHENOLS WITH ANTIOXIDANT, ANTI-ELASTASE, ANTI-COLLAGENASE, AND ANTI-TYROSINASE ACTIVITIES FROM INDIAN HIMALAYAN REGION

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Abstract: *Dendrobenthamidia capitata* (Wall.) Hutch. fruit (Bhamora) is a popular yet underexplored wild edible fruit of Uttarakhand state of Indian Himalayan region. Therefore, the present study was aimed to analyze *D. capitata* fruit for its phenolic contents, antioxidant, anti-elastase, anti-collagenase, and anti-tyrosinase activities using in- vitro assays. Phenolics and flavonoids were extracted using 80 % aqueous acetone and acid acetone as solvents. Our analysis revealed high values of total phenolic contents of 4.38±0.08 mg gallic acid equivalents (GAE). g^{-1} fruit weight and total flavonoid contents of 7.47±0.66 mg catechin equivalents (CE). g^{-1} fruit weight in the acid acetone fruit extracts of *D. capitata. D. capitata* fruit extracts demonstrated remarkable antioxidant activities (ABTS+, 1,1-diphenyl-2-picrylhydrazyl, superoxide anion, linoleate peroxyl radicals scavenging, and ferric reducing activities) and notable anti-elastase, anti-collagenase, and anti-tyrosinase activities. High resolution liquid chromatography–mass spectroscopy analysis revealed presence of six phenolic compounds, namely, dihydrorobinetin, dhydromyricetin, retusin dimethyl ether, rotenone, bergenin (C-glycoside of 4-O-methyl gallic acid) and peucenin in fruit extracts. This study recommends utilization of *D.capitata* fruit as functional food with prospective pharmaceutical, nutraceutical, and cosmeceutical properties.

Keywords: Wild edible fruits, *D. capitata*, Antioxidant, Anti-elastase, Anti-collagenase, Anti-tyrosinase

Introduction

Increased consumption of vegetables and fruits has been acknowledged to be associated with lower risk of a number of chronic diseases, such as coronary heart diseases, cancers, immune dysfunction and diabetes owing to their unique combination of minerals, vitamins and polyphenols (Pandey and Rizvi, 2009). Recently, fruits have attracted tremendous attention among food scientists, nutritionists and consumers owing to their multiple health promoting properties including antioxidant, antimicrobial, anticancerous, antidiabetic, anti-inflammatory, anti-skin aging and anti-skin darkening activities associated with elevated polyphenol contents and delicious taste respectively (Wang et al., 1996; Sun et al., 2002; Mathur et al., 2011; Mueller et al., 2010; Dai and Mumper, 2010; Li et al., 2014; Mukherjee et al., 2011; Hu 2011; Chang 2009). Furthermore to cultivated fruits, wild edible fruits have also gained global concern due to their higher polyphenol contents and outstanding antioxidant activities (Meda et al., 2008; Chalise et al., 2010; Fu et al., 2010; Murillo et al. 2012; Siqueira et al., 2013). In continuation to the worldwide research, wild edible fruits of Indian Himalayan region (IHR) have also emerged as potential source of nutraceuticals including polyphenols with antioxidant, anticancer, anti-

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THE STUDY OF GROWTH KINETICS OF *BACILLUS SUBTILIS* BMT4I (MTCC 9447) USING MOBIL OIL AS THE SOLE CARBON AND ENERGY SOURCE

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Abstract: The present study demonstrated the growth kinetics of a potent high molecular weight polycyclic aromatic hydrocarbon (HMW-PAH) degrader *Bacillus subtilis* BMT4i (MTCC 9447) using mobil oil or engine oil as the sole carbon and energy source. The study was performed to demonstrate the variations in morphology and growth kinetics in benzo-a-pyrene (BaP) degrading BMT4i due to the mobil oil induced stress conditions. The morphological variations were evaluated using Gram staining and spore staining (Schaeffer-Fulton method) followed by assessment of viability and growth using colony forming units (CFU)/ml method by means of growing *Bacillus subtilis* BMT4i in basal salt medium (BSM) with mobil oil (2%: BSMM) as sole carbon and energy source at different time intervals. The findings represented that mobil oil has noticeable effect on the shape and size of BMT4i cells. After 24 h of exposure to mobil oil, maximum BMT4i cells entered in to endospore development and several exospores were released after 24 h. The growth kinetics depicted an exponential increase in BMT4i cells in BSMM with increase in incubation time up to 3 days accomplishing maxima of 2.1 x 10^{19} demonstrating approx. 2x10^{11}-fold enhancements in cell number and afterwards cell number declined. Increase in CFU number (approx. 2x10^{11} fold) was directly linked with the BMT4i potential to use mobil oil as the sole carbon and energy source leading to elevation in cell number within just 3 days. Thus, it could be concluded that *Bacillus subtilis* BMT4i (MTCC 9447) is very competent in using mobil oil as the sole source of carbon and energy and hence it could be employed for bioremediation of mobil oil and PAH contaminated sites.

Keywords: *Bacillus subtilis* BMT4i (MTCC 9447), Benzo-a-pyrene (BaP), Bioremediation, Degradation, Mobil oil.

Introduction

Mobil oil or Engine oil is the oil which is used for lubrication of various internal combustion engines and it is one of the several refined products or cuts of crude oil (Bagherzadeh-Namazi et al., 2008). It is composed of a mixture of base polycyclic aromatic hydrocarbons (PAHs) having saturated and unsaturated long-chain with C16–C36 carbon length, cyclic alkanes and additives such as anticorrosive, antiwearing and antitearing agents (Koma et al., 2003). The main function is to lubricate moving parts; it also cleans, inhibits corrosion, improves sealing, and cools the engine by carrying heat away from the moving parts (Hagwell et al., 1992). The most important characteristic of the lubricating oil for automotive use is its viscosity (Tripathi and Vinu, 2015). The illegal dumping of used engine oil is dangerous to the environment and constitutes a serious threat to humans, animals, and vegetation (Bhattacharya and Biswas, 2014). After sometime of usage in engine operation, the lubricating oil become enriched with more metals and heavy polycyclic aromatic hydrocarbons (PAHs) that could pose chronic hazard including mutagenicity and carcinogenicity (Wong and Wang, 2001; Dominguez-Rusado et al., 2003; Adelowo et al., 2006; Lu S-T et al., 2008).

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Original Research Article

Biotechnology for health care

Holoptelea Integrifolia Bark And Leaves Phytochemicals, An Effective Ethno-Pharmacological Remedy For Treating Oral-Dental Infections

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Abstract: *Holoptelea integrifolia* (Ulmaceae) is a versatile medicinal plant encompassing a wide range of pharmacological properties. The present study is aimed at assessment of anti-bacterial activities of bark and leaf phytochemicals against the common oral bacterial pathogens namely *Staphylococcus aureus, Streptococcus oralis, Streptococcus mutans, Streptococcus gordonii, Lactobacillus brevis* and *Lactobacillus casei*. Bark and leaf dried powder samples were Soxhlet fractionated with solvents viz. petroleum-ether, chloroform, absolute-ethanol, 80%-methanol and distilled-water. Antibacterial potentials of the fractionated extracts against oral pathogens were evaluated with respect to zone of inhibition, MIC, MBC and IC$_{50}$ values whereas antioxidant activities were assessed against ABTS$^+$ and DPPH free radicals. The results demonstrated significantly higher antibacterial activity in bark ethanolic extracts (HiBE) towards every tested oral bacterial pathogen as established by its greater zone of inhibition and lowest MIC, MBC and IC$_{50}$ values. HiBE most potent antibacterial action against *L. casei* followed by *L. brevis, S. mutans, S. aureus, S. gordonii and S. oralis* HiBE showed remarkable ABTS$^+$ and DPPH free radicals scavenging activities with abundance of phenolics, flavanoids and tannins. Therefore, the present study ascertains *H. integrifolia* bark extracts as a commendable natural remedy for treating oral-dental infections.

Keywords: Antibacterial activity; antioxidant; *H. integrifolia* bark and leaves; oral bacterial pathogens; polyphenols; oral diseases

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ANTI-CARIOGENIC POTENTIALS OF *VITEX NEGUNDO* LINN

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Abstract: *Vitex negundo* Linn. (*Verbenaceae*), is an ethnobotanical important shrub carrying a large array of pharmacologically active phytochemicals with diverse medicinal properties. Present study assessed the antibacterial activities of leaf and twig (without leaf) bioactives against common oral bacterial inhabitants viz., *Staphylococcus aureus, Streptococcus oralis, Streptococcus gordonii, Streptococcus mutans, Lactobacillus brevis* and *Lactobacillus casei*. Leaf and twig dried powdered samples of *V. negundo* were separately fractionated by five solvents in eluotropic series namely petroleum-ether, chloroform, absolute-ethanol, 80%-methanol and distilled-water. Extracts were further analyzed for total phenolic contents (TPC), flavonoids (TFC), tannins, alkaloids and terpenoids. Zone of inhibition, MIC, MBC and IC_{50} values of the fractionated extracts against oral bacterial pathogens were determined. The results demonstrated significantly higher antibacterial activity in ethanolic extracts (VnLEt) and chloroform (VnLCh) extracts of leaves as evident by their higher ZOI and lower MIC, MBC and IC_{50} values. VnLEt extracts showed bactericidal effects against all the tested six bacterial pathogens (*L. casei>St. gordonii>Staphylococcus aureus>L. brevis>St. oralis=St. mutans*). VnLCh showed notable bactericidal effects against five pathogens (*L. casei>St. mutans>St. gordonii>Staphylococcus aureus*) but remained ineffective against *St. oralis* and *L. brevis*. Among twig extracts, ethanolic extracts showed inhibition against *Staphylococcus aureus, St. gordonii* and *St. mutans* while chloroform extracts showed antibacterial effect only against *Staphylococcus aureus*. VnLEt extract showed abundance in TPC, TFC and tannins while VnLCh was rich in alkaloids and terpenoid suggesting their contribution in bactericidal effects against cariogenic bacterial pathogens. Hence, the present study successfully established *V. negundo* leaf extracts as an excellent natural remedy for oral-dental infections.

Keywords: Antibacterial activity, *Vitex negundo,* Oral bacterial pathogens; Oral diseases

Introduction

Maintenance of oral hygiene is very important to prevent oral ailments including dental caries and periodontal disorders. More than 750 microbial species are known to reside in oral cavities and many of them have been documented in contributing to oral diseases (Jenkinson et al. 2005). *Streptococcus* species (*Streptococcus mutans, St. sobrinus, St. oralis* and *St. gordonii*), *Pseudomonas* species (*P. aeruginosa* and *P. fluorescens*), *Staphylococcus* species (*Staphylococcus aureus* and *S. epidermidis*), *Lactobacillus* (*L. brevis* and *L. casei*) and *actinomycetes* and yeast, mainly *Actinomyces*, *Actinobacillus* and *Candida* species, are the chief oral inhabitants responsible for oral infections leading to initiation and progression of dental caries (Tanzer and Livingston 2001; Karpiński 2013; Jamal et al. 2018). While, a variety of drugs including dentifrices, antiseptics and antibiotics are being utilized for the management of oral infections however, they produce several undesirable side effects including tooth coloring,

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Comparative Assessment Of Antioxidant And Antiproliferative Activities Of Pulp And Seed Of Himalayan Bayberry Fruit

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Abstract: *Myrica esculenta* Buch-ham. Ex D. Don or "Himalayan bayberry" (family *Myricaceae*) is an extremely valuable wild edible plant whose root, bark, leaves and fruits possess therapeutic properties and are active ingredient in several ayurvedic formulations employed for the treatment of various ailments and disorders including asthma, chronic bronchitis, ulcers, anemia, fever, diarrhea etc. Polyphenols from Himalayan bayberry fruit pulp and seeds were extracted using 80% acetone. TPC and TFC in the pulp and seed extracts were determined according to the Folin-Ciocalteau method and aluminum chloride method respectively. Antioxidant potentials of pulp and seed extracts were determined as DPPHRSA, ABTSRSA and FRA. Anti-proliferative activities of the fruit pulp and seed extracts were analyzed against breast cancer cell line MDA-MB231. 80% acetone extracts of seed (MeSA) showed very high TPC (4143.02±29.3 mg Gallic acid equivalent/100 g FW and TFC (1397.72±28.44 mg catechin equivalent/100 FW) which were 11 and 5 times greater than that were found in pulp extracts (MePA). MeSA extracts showed exceptionally high antioxidant activities with respect to the DPPHRSA and ABTSRSA (IC_{50}, 0.1153 mg/ml and 0.0017 mg/ml respectively) and ferric reducing activities. In addition, MeSA extracts was highly toxic to the breast cancer cell line MDA-MB231 showing 92% inhibition at 10 mg/ml while remaining ineffective against normal transformed cell line HEK293. The inhibition was much higher than that showed by pulp extracts (64%) at the same concentrations. This is the first report demonstrating an excellent ability of *M. esculenta* seed aqueous acetone extract with superior antioxidant activity and antiproliferative potential against breast cancer cells. Therefore, this study recommends utilization of *M. esculenta* seed for the development of nutraceutical and pharmaceutical formulations.

Keywords: Antioxidant; Antiproliferative; Flavonoids; Himalayan bayberry; *Myrica esculenta*; Phenolics

Introduction

Myrica esculenta, Buch-ham. Ex D. Don (family *Myricaceae*), commonly known as "Kaphal", or "Himalayan bayberry" is amongst highly valued wild edible plants growing between 900 and 2100 m above sea level distributed from Ravi eastward to Assam, Khasi, Jaintia, Naga and Lunshi hills in Indian Himalayan Region and extended to Malaya, Singapore, China and Japan (Gaur 2000). Himalayan bayberry is an evergreen tree, up to 14 m high, bark brownish-grey, rough, vertically wrinkled. Leaves alternate, crowded at the end of branches, oblanceolate, 6-15x3-5 cm, entire, acute, glossy above, glaucous dotted below, petioles 7-14 mm long. *M.esculenta* flowering and fruiting season is August to October and April to June respectively (Gaur 2000). Different parts of the *M. esculenta* plant including root, bark, leaves and fruits possess therapeutic properties and are part of numerous ayurvedic formulations used for treatment of various ailments and disorders such as asthma, cough, chronic bronchitis, ulcers, inflammation, anemia, fever, diarrhea, and ear, nose and throat

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Short Communication Open Access

Association between *IL6* gene polymorphism and the risk of chronic obstructive pulmonary disease in the north Indian population

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ABSTRACT

Interleukin-6 (IL6) is encoded by the *IL6* gene in human and acts as pro-inflammatory cytokine and an anti-inflammatory cytokine. Recent studies established that *IL6* substantially contribute in the diagnosed of systemic inflammation for the patients suffering from lung diseases such as chronic obstructive pulmonary disease (COPD). Thereof, this work aimed to investigate the protagonist of *IL6* (-174 G/C) genotypes as an essential risk factor for COPD in north Indian population. In the study, a total of 200 clinically diagnosed patients with COPD were selected against 200 patients. Statistical analysis reveleaed that there was no significant association between the *IL6* -174 G/C genetic polymorphism and the risk of COPD (P>0.05).

Keywords: Chronic obstructive pulmonary disease; Interleukin-6; Genotypes; Cytokine

INTRODUCTION

Chronic obstructive pulmonary disease (COPD) has been categorised as systemic disease and characterized for narrowing or obstruction of airways and resulted into chronic bronchitis or emphysema. It has been also considered with abnormal inflammation in the respiratory tract and lung by noxious particles or gases [1]. Generally, smoking has been suggested as major factor for inducing COPD risk but specific mechanism for the pathogenesis is not adequately studied [2]. Recent studies suggested that individuals diagnosed for only chronic bronchitis with no substantive airflow limitations should not be characterized as COPD 3]. Moreover, recent investigations also established the COPD association with smoking as symptomatic COPD development in 50% smokers [4].

Cytokines are major components for chronic inflammation in all the diseases as well as in COPD [5]. It was reported that *IL6* gene (MIM:147620) plays an active role in pathogenesis of lung disease like asthma [6]. Hence, *IL6* gene may be a relevant and appropriate target for the treatment of COPD and other associated chronic lung diseases [7]. The present study was conducted to establish the relationship between *IL6* (-174 G/C) polymorphism and risk of COPD in a population from the northern region part of India.

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Short Communication Open Access

Insertion/deletion polymorphism of angiotensin-converting enzyme and chronic obstructive pulmonary disease: A case-control study on north Indian population

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ABSTRACT

This research aimed to explore the *ACE* (insertion/deletion) gene association as key factor for chronic obstructive pulmonary disease (COPD) development in north Indian population. A total of 200 clinically diagnosed patients with COPD were selected against 200 healthy individuals. Genetic variations of *ACE* (insertion/deletion) were evaluated by using polymerase chain reaction techniques. Smoker showed higher risk of COPD (OR=1.67, 95% CI=1.12-2.48, P=0.012). Present results revealed the positive association between the DD genotype and the risk of COPD (OR= 2.14, 95% CI=1.22-3.78, P=0.006). Among smokers, DD genotype showed statistically significant association with increased risk of COPD (OR=3.10, 95% CI= 1.50-6.47, P=0.001).

Keywords: ACE; COPD; Polymorphism; Genotype

INTRODUCTION

Chronic obstructive pulmonary disease (COPD), is defined as an inflammation in the respiratory tract. In addition to participation of metabolic and cytokine gene in COPD development, *ACE* (Angiotensin-Converting Enzyme) gene was also suggested as essential factor responsible for the progression of pulmonary hypertension which may leads to COPD [1].

The *ACE* gene which is located at chromosome 17 (17q23 region) was discovered with insertion I allele or deletion D allele polymorphism of 287 base-pair in *Alu* repeats nonsense DNA domain [2]. The association between *ACE* DD genotype and pulmonary hypertension during exercise in COPD patients has been reported. This excessive pulmonary hypertension due to exercise was suggested in the disturbance of oxygen passage into the tissues. Significant co-relation between the *ACE* DD genotype and COPD risk with pulmonary hypertension amongst the Asian people by meta-analysis was established [3]. The involvement of renin in angiotensinogen conversion into angiotensin I was suggested as the first part for renin-angiotensin system (RAS) activation [4]. However, association of the renin-angiotensin system (RAS) with COPD pathophysiology was also documented. Moreover, retrospective studies reported that ACE (angiotensin converting enzyme) inhibitors and RAS-reduce the mortality in COPD patients. Thereof, it is seen as promising therapeutic approach for COPD [5, 6]. In this

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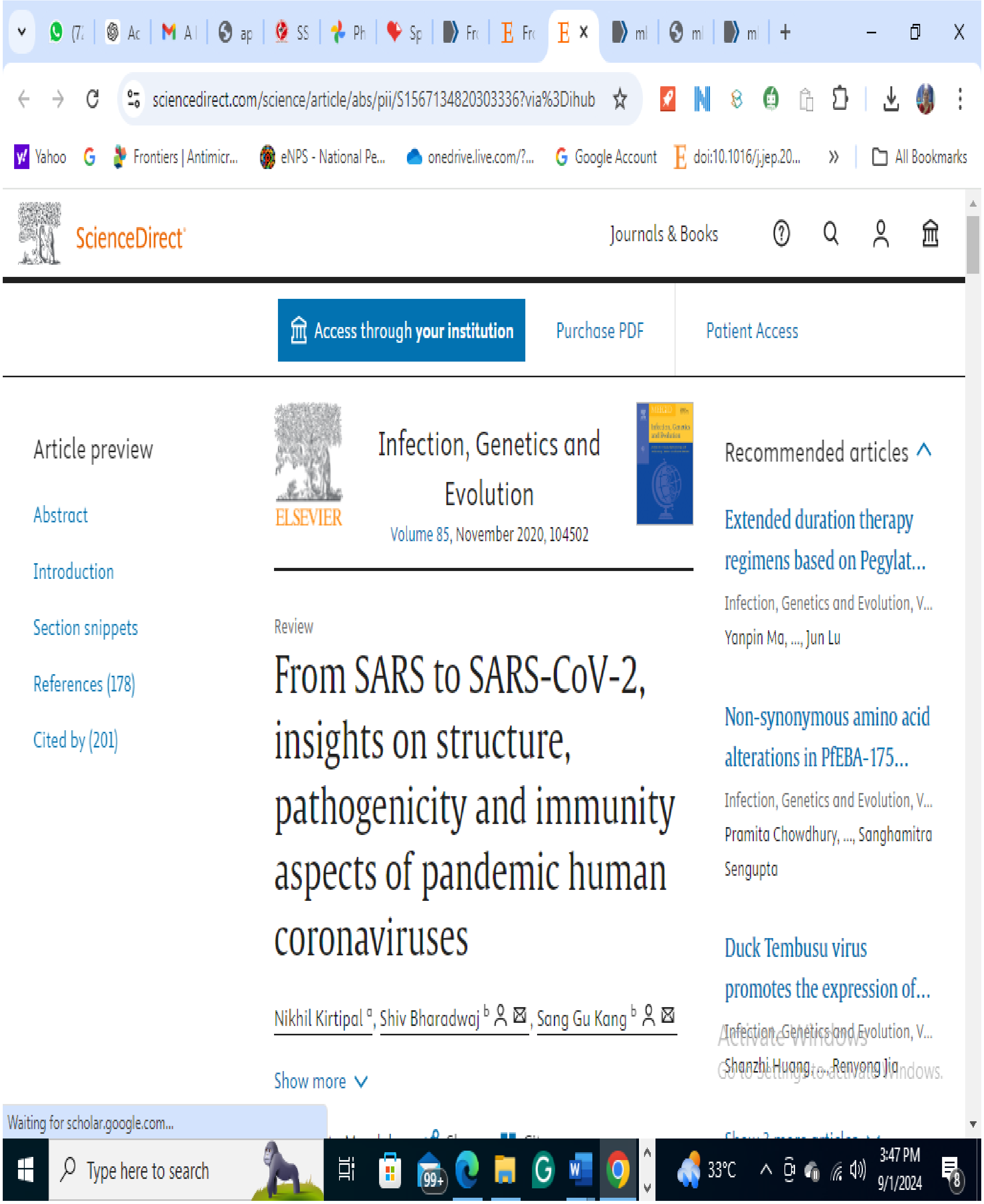

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Review

From SARS to SARS-CoV-2, insights on structure, pathogenicity and immunity aspects of pandemic human coronaviruses

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Benzo (A) Pyrene Detoxification Potential of *Bacillus subtilis* BMT4i (MTCC 9447) Isolated From Srinagar, Garhwal, Uttarakhand, India

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Abstracts: Benzo(a)Pyrene (BaP), a pentacyclic high molecular weight polyaromatic hydrocarbon (HMWPAH) is a priority pollutant of extreme environmental concern due to its potent carcinogenic properties. *Bacillus subtilis* BMT4i (MTCC 9447) a well known BaP degrader may play a major role in the bioremediation of contaminated soils. Using multiple pathways, BMT4i efficiently degrades BaP into several metabolites, whose mutagenicity remains unknown. Present investigation aimed to analyze the BaP detoxification potential of BMT4i by evaluating the mutagenicity of BaP metabolites using the Ames test. For that, TA98 strain of *Salmonella typhimurium* (*his*D3052 mutant) was incubated with ethyl-acetate extracts of 7, 15 and 40 days grown cultures of BaP-BMT4i with or without S9 activation in the medium devoid of Histidine. The number of yellow revertant colonies representing mutation was counted and % detoxification potential was evaluated. The results showed that in the absence of S9 activation, a very small comparable number of revertant TA98 colonies were observed in negative control and test samples ($p>0.05$). After S9 activation a large number of revertant TA98 colonies (43±/48) appeared in negative control (BaP standard) which significantly reduced ($p< 0.001$) after 7 (23±3/48), 15 (12.4±2.7/48) and 40 days (8.3+1.9/48) demonstrating decrease in the mutagenicity of BaP by BMT4i. The percent BaP detoxification after 7 days was 45% which further increased to 70 and 80% after 15 and 40 days. The commendable in-vitro BaP detoxification by *Bacillus subtilis* BMT4i needs to be further tested through field trials in order to establish it as an efficient bio-weapon in bioremediation of HMW-PAHs contaminated sites.

Keywords: Ames test • *Bacillus subtilis* BMT4i • Benzo(a)Pyrene (BaP) degradation • BaP detoxification • mutagenicity

Introduction

Benzo(a)Pyrene (BaP), a five ring high molecular weight polyaromatic hydrocarbon (HMWPAH) is a potent carcinogen with high recalcitrant nature. It is released into the environment primarily by anthropogenic activities including incomplete combustion of fossil fuel, coke oven emissions, coal combustion, conversion industries, aluminum smelters, vehicle exhausts, cigarette, cigar and marijuana smoking (Kanaly and Harayama 2010). The natural sources of BaP include forest fires, peat fires, volcanoes, burning of crude oil and shale oil (Juhasz and Naidu 2000). From contaminated soil and water, BaP enters into the food chain and is exposed to humans. In addition, ingestion of foods cooked over an open flame or smoked (Saito et al. 1978) and use of cosmetics which contain mineral oil (Grob et al. 1991) resulted in BaP exposure. In the liver, BaP is metabolized into its genotoxic form, which then interacts with nucleic acids and proteins, creating highly reactive macromolecular adducts. This leads to BaP-induced toxicity, mutagenesis, and carcinogenesis (IARC 2010, EPA 2017). Due to its abundance in the

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Herbal Pesticides, a Natural Weapon to Control the Insect Pests: A Review

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Abstract: Agro inputs are increasing globally day by day by the farmers. However, production, its quality and productivity are not increasing proportionate to incremental expenditure on farming; thus causing agriculture becoming more and more unviable proposition in today's context. Synthetic fertilizers and pesticides are causing unbearable damage to ecology and environment and agriculture cannot sustain on continuous use of such types of fertilizers and pesticides. Synthetic pesticides have been used to control the insects since a long time. This practice gradually affected on the quality and quantity of plants by degrading them. The use of synthetic pesticides on small-scale farms is generally not advocated because the approach lacks sustainability and raises environmental and health concerns. These synthetic pesticides not only affect the negative impact on plants but many farmers simply cannot afford the cost of such pesticides. Therefore, it is high time to screen for safe and effective biodegradable pesticides which is cost-effective with non-toxic effects on non-target organisms. The present review includes the prospects and utilization of plant extracts as natural weapon against various insects.

Keywords: Herbal pesticides, Synthetic pesticides, Natural weapon

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Farmers are generally dependent on using synthetic pesticides to control the plants from damage caused by insects (Rahman *et al.*, 2007). The repeated use of synthetic insecticides for insect pests and vector control has disrupted natural biological control systems. It has also resulted in the development of resistance, undesirable effects on non-target organisms and fostered environmental and human health concerns, which initiated a search for alternative control measures (Adeniyi *et al.*, 2010). Most plant

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Circulatory System of *Columbicola columbae* Linnaeus (Phthiraptera: Insecta: Ischnocera: Philopteridae) Infesting Blue Rock Pigeon (*Columba livia* Gmelin)

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Abstract: Circulatory system of pigeon slender louse, *Columbicola columbae* (Phthiraptera: Ischnocera) has been studied in greater details. In arthropodan insects, the circulatory system is typically open, with just one closed artery. The heart of *C. columbae* is a straightforward, elongated, and single-chambered organ. At its anterior, middle, and basal regions, it has three pairs of laterally positioned minute ostia. From the body wall to the heart, six sets of alariform muscles—three on the left and three on the right—extend in a fan-like pattern.

Keywords: Circulatory system • *Columbicola columbae* • Phthiraptera • Ischnocera

Introduction

The number and types of haemocytes, developmental stage and physiological state vary with insect species. Haemocytes are blood cells that circulate in a clear fluid, the plasma, within the haemocoel (body cavity) of insects. Seguy (1951) and Eichler (1963) have reviewed the work done on anatomical peculiarities of some mallophagan lice. Anatomy of different organ systems of two amblyceran Phthiraptera *Laemobothrion percnopteri* (Srivastava, 1974) and *Menacanthus eurysternus* (Chandra, 1986) and one ischnoceran poultry louse, *Lipeurus lawrensis tropicalis* (Saxena, 1979) have been studied in greater details.

Morphological features of cerebral and suboesophageal ganglion of *Trimenopon jenninysi* (Stowe, 1943), *Bocivola caprae* (Risler, 1951), *Pseudomenopon pilosum* and *Ornithobius cygni* (Haub, 1967 & 1971) have already been studied. Specific information on digestive and tracheal system of few selected phthirapteran species has been contributed by Haug (1952), Waterhouse (1953), Saxena & Agarwal (1981), Navioe*t al.*, (1986), Arya & Singh (2021) and Singh & Arya (2022). The nature of heart and aorta of few species has been noted by Fulmek (1906) and Saxena & Agarwal (1980a). Only few papers Mayer (1954), Lawrence (1956), Saxena & Agarwal, (1979 & 80b), Agarwal & Saxena, (1981); Singh *et al.*, (1985), Ribeiro & Brehelin (2006), Symmons & Hindle (2010) and Arya (2020) deal with blood cells or circulatory system of phthirapteran ectoparasites.

Workers like, Wigglesworth (1955 & 59), Jones (1962), Vostal (1969), Arnold (1974) and Gupta (1979) have discussed the nature and classification of insect haemocytes.

The present study deals with circulatory system of an ischnoceran pigeon louse, *Columbicola columbae* in greater details.

Material and Methods

Fresh lice were collected directly from experimental hosts (pigeon). In addition, regular supply of lice was secured by maintaining them *in vitro* condition at 35±1°C temperature, 75–90% Relative Humidity and pinfeathers for feeding in incubator. Fine sharp entomological pins (after fabricated by rubbing on fine sandpaper) were used for successful dissection. Both the sexes of lice were dissected under a stereozoom binocular

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DIGESTIVE AND TRACHEAL SYSTEM OF *COLUMBICOLA COLUMBAE* LINNAEUS (PHTHIRAPTERA: INSECTA: ISCHNOCERA: PHILOPTERIDAE) INFESTING BLUE ROCK PIGEON (*COLUMBA LIVIA* GMELIN)

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Abstract - Anatomical aspects (digestive and tracheal system) of an ischnceran pigeon slender louse, Columbicola columbae (Phthiraptera) has been studied in greater details. Alimentary canal of C. columbae consists of three usual regions, fore-gut, mid-gut and hind-gut. The crop is blind tadpole shaped structure and its anterior end is beset with 25-35 groups of cropteeth, each containing 4-10 teeth, measuring 2.5 – 5.1µm in length. Four long blind Malphighian tubules (each divisible into three portion) arises from junction of mid-gut and hind-gut. The nature of Malphighian tubules remains similar in two sexes but diameter of gut is narrower in males, in comparison to females. Salivary apparatus consists of a pair of rounded ball like salivary gland, a pair of spherical reservoir and their ducts. Six small apertures like stigmata occur on the lateral margins of abdominal segments of C. columbae. A pair of spiracle also occurs in pterothorax, near the base of third pair of legs. Tracheae are disposed in two main lateral longitudinal trunks which are connected to each other in pro-, meso- metathorax and in last abdominal segment. Branches arising from these longitudinal trunks in each segment tracheate various muscles and visceral organs.

Keywords: Pigeon louse, Columbicola columbae; Phthiraptera; Ischnocera; Digestive system; Tracheal system.

1. INTRODUCTION

Considerable preliminary information on anatomy of phthirapteran ectoparasites have been provided by Nitzsch, 1818; Kramer, 1869; Nusbaum, 1882; Grosse, 1885; Snodgrass, 1899; Gross, 1905; Shipley, 1909; Strindberg, 1916a, 1916b, 1918 & 1919; Blagovestchensky, 1931 & 1959 and Clay, 1949. Seguy (1951) and Eichler (1963) have reviewed the available information on anatomical peculiarities of some lice. Anatomical aspects of different organ systems of three phthirapteran species [one Ischnocera, *Lipeurus lawrensis tropicalis* (Saxena, 1979) and two Amblycera, *Laemobothrion percnopteri* (Srivastava, 1974) and *Menacanthus eurysternus* (Chandra, 1986)] have already been worked out.

Specific information on digestive system of few selected species has contributed by Haug (1952), Waterhouse (1953), Saxena & Agarwal (1981) and Navio *et al.*, (1986). Wilson (1933), Crutchfield & Hixson (1943), Agarwal *et al.*, (1983), Saxena *et al.*, (1985), Trivedi *et al.*, (1990), Singh *et al.*, (2010) and Kumar *et al.*, (2017) added valuable information about haematophagous nature and feeding habits of some amblyceran and ischnoceran avian lice species. Publications on structure and role of cropteeth of some Mallophaga have also appeared from time to time (Cummings, 1913; Blagovestchensky, 1949; Agarwal & Saxena, 1978; Saxena & Agarwal, 1979; Srivastava & Agarwal, 1981; Saxena *et al.*, 1995). Recently, circulatory system (Arya, 2020) and digestive and tracheal system (Arya & Singh, 2021) of poultry shaft louse, *Menopon gallinae* has been studied in greater details.

The present report deals with morphological features of digestive and tracheal system of an Ischnoceran pigeon slender louse, *Columbicola columbae*.

2. MATERIAL AND METHODS

Several fresh lice were collected directly from experimental hosts (pigeon). In addition, regular supply of lice was secured by maintaining them *in vitro* condition at 35±1°C temperature, 75–90% Relative Humidity and pinfeathers for feeding in incubator. Fine sharp entomological pins (after fabricated by rubbing on fine sand paper) were used for successful dissection. Both the sexes of lice were dissected under a stereozoom

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Digestive And Tracheal System of *Menopon Gallinae* (Phthiraptera: Amblycera) Infesting Poultry Bird (*Gallus Gallus Domesticus*)

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Abstract: The digestive and tracheal system of a poultry shaft louse, *Menopon gallinae* (Phthiraptera: Amblycera) has been studied in greater details. Alimentary canal of louse was dissected out along with crop under stereozoom binocular microscope. Entire alimentary canal of *M. gallinae* was found more or less straight and has three basic parts (fore-gut, mid-gut and hind-gut) while crop-teeth was present in the crop. The posterior end of crop contains 20-30 well developed crop-teeth arranged in a single arced plate, in comb-like fashion. Mid-gut was found as simple tube and contributes nearly one half of the total length, while the hind-gut was marked by the opening of Malpighian tubules in alimentary canal. The heart of *M. gallinae* is of simplest kind, one chambered bulbous structure having three pairs of laterally placed ostia and supported by four pairs of alary muscles. In the tracheal system there were seven pairs of spiracles occurred on the terga of *M. gallinae*. The first pair of spiracle was found located close to legs while remaining six abdominal spiracles occur from segment 3rd to 8th. The degree of tracheation of various visceral organs has also been noted.

Keywords: *Menopon gallinae* • Poultry shaft louse • Amblycera • Phthiraptera • Digestive system• Tracheal system

Introduction

Preliminary information on anatomy of lice (Phthiraptera) has been provided by Strindberg (1916a, 1916b, 1918 & 1919), Blagovestchensky (1931 & 1959) and Clay (1949), while Seguy (1951) and Eichler (1963) have tried to generalize the available information on anatomical peculiarities of some lice. Anatomical features of different organ systems of three species [one Ischnocera (*Lipeurus lawrensis tropicalis*) and two Amblycera (*Laemobothrion percnopteri* and *Menacanthus eurysternus*)] have already been worked out (Srivastava, 1974; Saxena, 1979; Chandra, 1986). Haug (1952), Waterhouse (1953), Saxena & Agarwal (1981) and Navio *et al.* (1986) have provided specific information on digestive system of selected species. Wilson (1933), Crutchfield & Hixson (1943), Agarwal *et al.* (1983), Saxena *et al.* (1985), Trivedi *et al.* (1990), Singh *et al.* (2010) and Kumar *et al.* (2017) added valuable information about haematophagous nature and feeding habits of some amblyceran and ischnoceran avian lice species. Some publications dealing with structure and role of cropteeth of Mallophaga have also appeared from time to time (Cummings, 1913; Blagovestchensky, 1949; Agarwal & Saxena, 1978; Saxena & Agarwal, 1979; Srivastava & Agarwal, 1981; Saxena *et al.*, 1995).

The present report deals with morphological features of digestive and tracheal system of an amblyceran poultry louse, *Menopon gallinae*.

Material and Methods

Fresh lice were obtained directly from hosts. Furthermore, regular supply of lice was ensured

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LETTER TO THE EDITOR

Interleukin 6 polymorphisms as an indicator of COVID-19 severity in humans

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To the Editor

Cytokines, small proteins (∼5-20 kDa) required in cell signaling and as immunomodulating agents in human body. Now, these cell signals are known to mediate many inflammatory processes in the lungs and associated with pathogenesis of a variety of respiratory disorders (C. S. Park et al., 2000; W. Y. Park et al., 2001; Shieh et al., 2019). Interleukin-6 (IL-6; glycoprotein of size 21 KDa), coded by *IL6* gene in human, acted as both an anti-inflammatory myokine and pro-inflammatory cytokine. Jin and Wang summarized the rationale and value of single nucleotide polymorphisms (SNPs) relation to immune responses, so-called "immunogenetic profiling" (Jin & Wang, 2003). Also, certain lung disease like chronic obstructive pulmonary disease (COPD) and asthma, disease severity was differentially observed via epidemiological and gene polymorphism analysis (Jin & Wang, 2003; Kirtipal et al., 2020). For instance, polymorphisms on the *IL6* promoter region are connected with the risk of pneumonia (Chen et al., 2015; Martinez-Ocaña et al., 2013; Solé-Violán et al., 2010) and *IL6* polymorphism such as -174 *G/C* SNP (He et al., 2009) and 321 *G/T* (Yanbaeva et al., 2009) were associated with COPD in different Caucasian population while in certain Asian population, it was not significantly associated with COPD (Kirtipal et al., 2020). Likewise, polymorphisms in the *IL6* was linked to certain viral infection like hepatitis C (HCV), influenza virus, and hepatitis B virus (HBV) (Linnik & Egli, 2016; Riazalhosseini et al., 2018). For instance, low-producing *IL6* genotype CC (*IL6* rs1800795 G174C) in patients was allied to unprompted clearance of HCV infection from contaminated blood products (Barrett et al., 2001). On contrary, high producing *IL6* genotype CC of rs1800795 174 G/C polymorphism, i.e. *GC* or *GG*, were associated with a greater likelihood of sustained virologic response (SVR) in patients coinfected with HCV and HIV (Nattermann et al., 2007). Also, lower IL-6 serum level and rs1800795-*IL6 CC* genotype in patients were marked to have an attenuated adoptive immune response against chronic HCV (Bogdanović et al., 2016). The *IL6*-174 *C/C* genotype also associated with respiratory syncytial virus (RSV) infection magnitude (Doyle et al., 2010). In current scenario of Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection, including symptoms of fever, cough, sore throat, runny nose and difficulty breathing (Boopathi, et al., 2020); defined as COVID-19 pandemic, a little is deciphered about IL-6 correlation and its polymorphism with pathogenesis of idiopathic pulmonary fibrosis (IPF) in SARS-CoV-2 infection, except its significant role to initiate the cytokine (IL-6) storm in SARS-CoV-2 infected patients. Additionally, SNPs in TMPRSS2 was suggested to influence the SARS-CoV2 entry into the cell (Paniri et al., 2020). As, IL-6 is generally considered to be a profibrotic molecule (Le et al., 2014; Moodley et al., 2003; O'Donoghue et al., 2012), an experimental study with the bleomycin model of pulmonary fibrosis suggested that inhibiting IL-6 in early phase of lung injury promotes fibrosis and that inhibition in the later stages of injury at the onset of fibrotic phase might ameliorate fibrosis (Kobayashi et al., 2015). Recently, it was demonstrated that *IL6* rs1800795 G allele could act as a protective factor while *IL10* rs1800896 A allele could act as a risk indicator in Pneumonia-induced sepsis in Chinese Han patients. Additionally, these *IL6* polymorphisms were associated with clinical stage of sepsis and have significant influence on the secretion of IL-6 and IL-10 in the patients. (Mao et al., 2017). A meta-analysis was also conducted to relate the *IL6* gene polymorphism with predisposition as well as disease severity of pneumonia, suggested the carrier status of *IL6* 174 C allele with higher IL-6 production and pneumonia severity (Ulhaq & Soraya, 2020). Under the given role of IL-6 as a central regulator for the CD4 T cell fate, *IL6* polymorphism can provide further insights on the COVID-19 pandemic. Hence, we suggested *IL6* gene polymorphisms as indicator to establish the severity or understand the pathology derived from COVID-19 infections in humans, including susceptibility of some individuals and having asymptomatic symptoms or else having immune to the infection as reported earlier in various diseases (Chen et al., 2015; Linnik & Egli, 2016; Martinez-Ocaña et al., 2013; Nattermann et al., 2007; Riazalhosseini et al., 2018; Solé-Violán et al., 2010). By combining information about the immunogenetic impact of *IL6* polymorphisms as reported earlier in lung diseases and viral diseases, we also advised to consider *IL6* polymorphism as a

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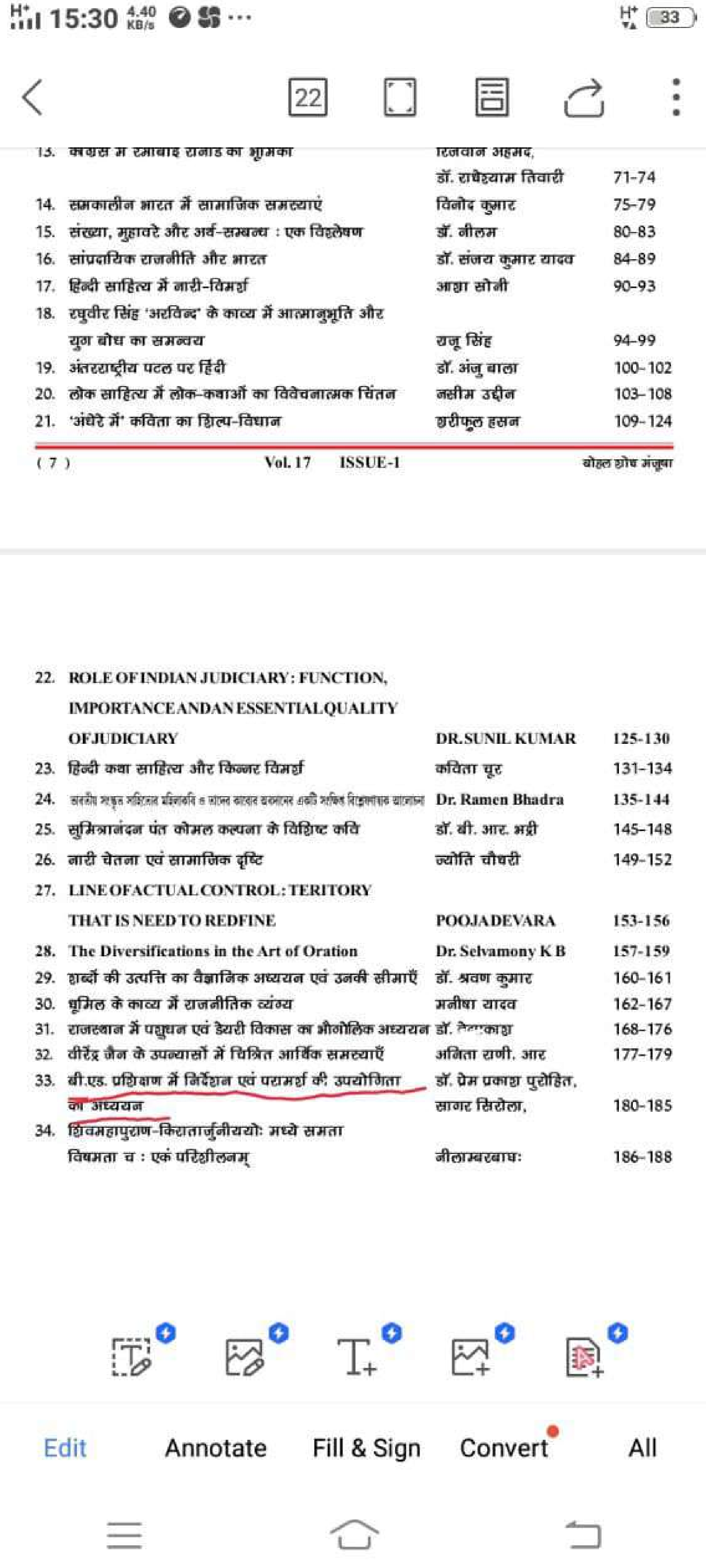
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