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Possible Prevention of COVID 19 by Using Linoleic Acid (C18) Rich Algae Oil

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ABSTRACT

With the rapid spread of COVID 19, people are being isolated in countries, and more than lakhs of people have been infecting by the coronavirus. The coronavirus is an airborne organism and highly infectious at short contact distances. The use of N95 respirators masks (high-efficiency) can protect people against the COVID 19, but the protective efficiency of masks is not high enough. A method of applying oil with rich amounts of linoleic acid in nostrils can prevent the spread of the virus. Macroalgal secondary metabolites have great potential for the development of new drugs and algae derived products largely employed in assorted industries, including agricultural, biomedical, food, and pharmaceutical industries. Among different chemical components isolated from algae, oil components are the most attracting more and which were subjected to a variety of studies (antiviral potential of algae in pharmaceutical research). Algal oil and their components like linoleic acid, oleic acid, palmitic acid, stearic acid are playing a preventive role in the virus infection. In addition, different mechanisms of action have been reported for these linoleic acid ($C_{18}H_{32}O_2$) components, such as inhibiting the binding virus into the host cells or suppressing virulence activity by destabilization of the bilayer of viral envelopes. Application of in controlling the virus entry is mainly depend on the properties like low surface tension, high-boiling point, high viscosity, immiscible with water and antivirus activity. This manuscript mainly discusses the possible physical-chemical mechanisms involved in the application of algal oil and other sources of oils component's role in prevention of viral spread. Among that we listed out various oil sources and their applications in controlling the virus activity. Further confirmed experimental and clinical results for the use of oils as nasal spray may finally contribute to preventing the spread of the coronavirus as soon as possible.

Keywords: Algal Oil in COVID 19 Control; Linolenic acid (C18) Coronavirus; COVID 19; Nasal spray, Algal Oil

1 Introduction

Algae are the undisputed primary producers in the aquatic ecosystem and contribute approximately half of the global net primary productivity. These photosynthetic organisms, including cyanobacteria abound in the planktonic region of the aquatic habitat are collectively called phytoplankton. These plankton communities together influence the global carbon cycle and ultimately the climate. Microalgae have been recognized as promising organisms for different metabolites production primarily due to their high growth rates compared with those of other photosynthetic organisms. These organisms has significant economic interest in oil as fuel, biochemical components as food and feed [Murrell et al., 2011]. Amongst natural products, higher amounts of biomedical compounds have been isolated from algae [Jha et al., 2004]. These organisms can synthetize assorted types of metabolisms, including fatty acids, amino acids, polysaccharides, chlorophyll, vitamins, xanthophylls, and halogenated compounds. Despite being underexploited plant resources, recent investigations have established algae as a rich arsenal of active metabolites with pharmaceutical potential, including anticancer, antitumor, antioxidant, antiobesity, neuroprotective, antimicrobial, antinociceptive, anti-inflammatory, and antiangiogenic activities [Moghadamtousi et al., 2014; Eom et al., 2012]. Metabolites produced from microalgae is considered as carbon neutral energy source. In contrast to other energy crops, microalgae can be cultivated on non- arable land. Besides, they can grow in marine or waste water and hence are not dependent on fresh water supply [Milano, J., et al 2016]. Water renewable rate for microalgae is much less than territorial plants. Many microalgal species display the ability to adapt and survive under varied environmental conditions and accumulate high lipids (tri-acyl glycerides (TAG)) under adverse conditions as a survival strategy to endure stress. Microalgae require low maintenance and have potential for high annual oil productivities. They can yield 40 times more oil per acre of land compared to other oil crops [Schenk, P.M., et al 2008]. The algal lipid production values reported in literature varies significantly; with as low as 245-gal acre-1 yr-1 (2.3 m^3 ha-1 yr-1) [Ramachandra, T.V., et al., 2013] to as high as 14,636-gal $acre^{-1}$ yr^{-1} (136.9 m^3 ha^{-1} yr^{-1}) [Mata, T.M., 2010]. Though lipid productivity of 5,000 gal $acre^{-1}$ yr^{-1} is considered as conservative value [Subhadra, B 2011], with currently achievable average annual algal biomass productivity of 14-16 g m^2 day^{-1} and existing harvesting and downstream technologies, production of about 1500-2000 gal acre-1 yr-1 seems to be a reality in scale-up open pond systems. Nonetheless, oil productivity from microalgae higher than many crops [Pienkos, P.T 2009] with good composition. Because of these above-mentioned reasons, microalgae gain significant importance in oil production.

Microalgae are creating interest in production of lipids either in the form of saturated or unsaturated and ω-3 fatty acids (FAs). Lipids from algal are considered as a source of n-3 polyunsaturated fatty acids (PUFAs) or as feedstock for the oil production and can be used as alternative sources of metabolic ingredients in functional food formulations. Algal lipid biochemistry has renewed interest to manipulate available renewable reservoirs using modern, advanced tools in biochemistry, and genetic engineering to solve many challenges to sustainably meet global challenges from food, energy to medicines. Algal lipids have unique compositions (higher levels of C18:3); in principle these compounds can be extracted and purified and can be used for different applications in pharmaceutical industry. They could potentially be used as nasal drops for controlling spread of coronaviruses. The source of saturation and unsaturated lipids from algae can act as an alternative source of medically important molecules for useful drug formulations.

In the present manuscript we are providing the possibility and possible mechanisms of linoleic acid (C18) and rich algal oil and the opportunities of C18:2 rich algal oils in prevention of 2019-nCoV.

2 COVID 19 Outbreak and Epidemiology

In December 2019, outbreak of the novel coronavirus disease caused by the coronavirus 2019-nCoV was now officially designated as severe acute respiratory syndrome-related coronavirus SARS-CoV-2. This virus is the seventh coronaviruses known to infect humans and is capable of human-to-human transmission. The viruses can cause respiratory syndrome-related infection symptoms and represents a pandemic threat to global public health [Zhu etal., 2020; Hui et al.,2019]. Morphologically, the 2019-nCoV is a spherical particle (60 nm to 140 nm under transmission electron microscope (TEM)), surrounded by 9-12 nm long viral spike peplomers (club-shaped), which are S-proteins located on virus envelope and determine host tropism [Zhu etal., 2020]. SARS-CoV-2 attacks the lower respiratory system to cause viral pneumonia. Viral S-proteins have strong binding affinity to human Angiotensin converting enzyme 2 (ACE2). When the S-proteins bind to the surface receptors of hose sensitive cells for further replication [Zhao, Yu, et al., 2020]. The ACE2 virus receptor expression is concentrated in a small population of type Ⅱ alveolar cells (AT2) in human lungs and the abundant expression of ACE2 in a population of AT2 leads to the severe alveolar damage namely pneumonia. [Zhao, Yu, et al., 2020]. SARS-CoV-2 may also affect the gastrointestinal system, heart, kidney, liver, and central nervous system leading to multiple organ failure. Current information indicates that SARSCoV- 2 is more transmissible/contagious than SARS-CoV. The 2019-nCoV is now rapidly spreading worldwide millions of people are being affected and isolated. Currently, the isolation of people is very necessary to control this viral pandemic.

Outbreak of the 2019-nCoV happened in winter and there were strong evidences of human-to-human transmission of 2019-nCoV among the close contacts of infected patients. Importantly, some of the 2019-nCoV-infected people are not showing any obvious clinical symptoms but were still capable of infecting other people in close contact and these asymptomatic infected people may pose a serious risk for the transmission of infection [Rothe, Camilla, et al., 2020; http://news.ifeng.com/c/7tg6pV2ymEE; https://tech.sina.com.cn/roll/2020-01-30/doc-iimxyqvy9139586.shtm]. Infection probability of COVID 19 is mostly influenced by the different factors. Among them environmental factors (Temperature, Air etc.,), personal hygienic and virulence of virus.

3 Possible Approaches in Prevention of COVID 19

Currently N95 respirators are being used as part of personal protective equipment (PPE), as well as surgical masks, gloves and goggles can offer a level of protection against the transmission of the coronavirus [Loeb, Mark, et al. 2004]. Some of the studies for the development of protective vaccines and drugs for COVID 19 is in under process [http://news.sina.com.cn/c/2020-01-26/doc-iihnzhha4771111.shtml].

3.1 Application of Oil in COVID 19 Control:

Some people, because of the fear about COVID 19, tend to use sesame oil as traditional medicine for preventing the infection. If sesame oil is added and coated to a person's nostrils, the person will not be infected by virus when the person exposed to patient or surrounding environments [https://www.2345daohang.com/dianji/64113/item_4521.html]. Recently, the Indian Government recommended the usage of sesame oil as drops in each nostril to prevent the new coronavirus according to their ancient ayurvedic medicine [https://pib.gov.in/PressReleasePage.aspx?PRID=1600895]. Scientifically there are some reasons to consider that the sesame oil as drops can provide protection to people from infection of virus. From the ancient days the people are using different sources of oils for body applications too. Mainly the origin of oils is from vegetable sources. The list of oils mostly belongs to the Canola (rapeseed) oil, Soybean oil, Pumpkin seed oil, Perilla seed oil, Tofu and Walnut oil. Present scenario as discussed in the introduction algal oil is could be a best renewable source for the medical usage and application. Algae gained momentum because of their ability in reducing carbon foot print through photosynthesis and cleaner energy.

3.2 Role of Algal and Production Strategies

i. Cultivation and Harvesting

Oil production from microalgae involves series of processes, which includes, cultivation, harvesting and downstream processing (Figure 1). Two most common methods of mass cultivation of microalgae are open systems like raceway or circular ponds and closed systems like flat plate or tubular Photo-Bio Reactors (PBRs). Open raceway ponds are most economical option for mass cultivation of algae because of their low cost of construction and maintenance [Chisti, Y., 2013]. Two most common methods of mass cultivation of microalgae are open systems like raceway or circular ponds and closed systems like flat plate or tubular Photo-Bio Reactors (PBRs). Open raceway ponds are most economical option for mass cultivation of algae because of their low cost of construction and maintenance [Chishti, Y., 2013].

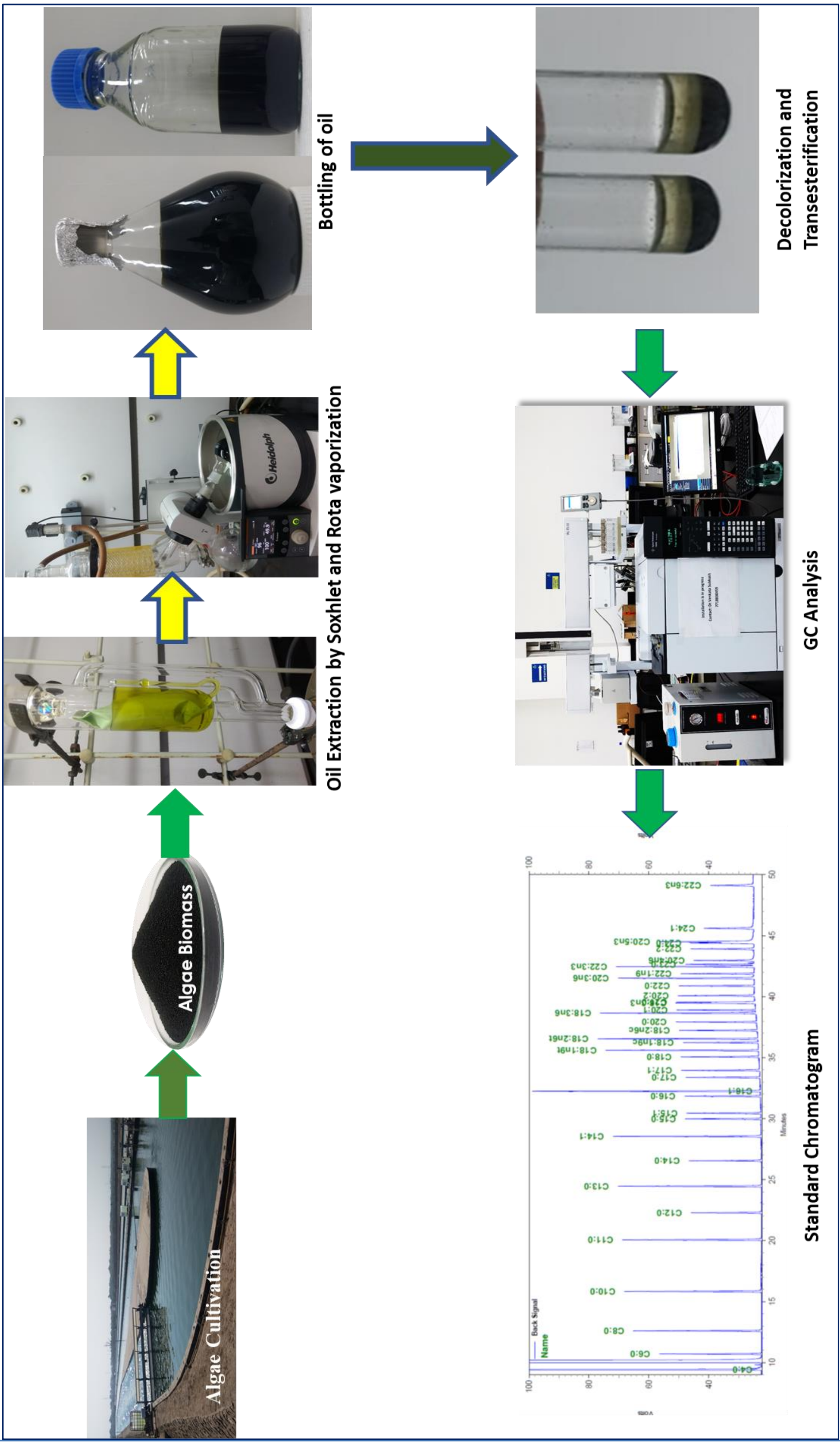

Fig 1: Steps involved in Oil production and compositional analysis

Selection of appropriate and robust algal strain is critical for success of metabolites production from microalgae. The algae strain must be resilient to withstand wide environmental fluctuations and grow all through the year with high biomass and lipid productivities. Furthermore, lipid profile governs the downstream extraction, conversion and stability of biodiesel. [Islam, M., et al., 2013]. Amongst several reported algal strains *Nannochloropsis sp.*, *Neochloris oleoabundans* and *Nannochloris sp.* are known to have high lipid content and biomass productivities and hence are potential strains for bio oil or bio-crude. Post cultivation is the recovery of microalgae from the cultivation medium. Both harvesting and dewatering are together considered as a single operation or as combination of sequential operations. Once microalgae are harvested and dewatered, the resulting biomass is either dried or used directly as feedstock for production of biocrude or other downstream processing. Microalgae can be dried using one of the several methods reported in the literature; such as spray-drying, drum-drying, fluidized bed drying, freeze-drying, refractance window dehydration technology, low-pressure shelf drying or sun-drying. Out of all, sun drying is most economical method but needs large drying area.

*ii.*Lipid Production and Types

Algal lipids are of two types namely, polar and non-polar lipids. Polar lipids, such as phosphoglycerides and glycosylglycerides are structural elements of algal membranes and play an important role in transport and cell signaling. On the other hand, non- polar lipids are storage lipids and help to cope with the stress conditions. Many microalgal species can produce large amount of triacylglycerols (TAG- storage lipid) under stress conditions, accounting for 20– 50% of the dry cell weight. Increase in TAG content in stressed or stationary phase cells is primarily due to metabolic shift from membrane lipid biosynthesis to storage lipids synthesis. As in microalgae, cyanobacteria do not have TAGs. Fatty acids (FA) are the building blocks for lipid synthesis and are primarily synthesized in the chloroplast. First committed step in fatty acid synthesis is conversion of acetyl CoA into malonyl CoA. After first committed step, malonyl group is transferred to protein co-factor on acyl protein carrier (ACP). Malonyl ACP is the carbon source for subsequent elongation reactions, forming saturated 16:0 and 18:0-ACP.

Algae are able to produce a wide variety of C12 to C24 carbon length fatty acids and their oxidized products (oxylipins), and sterols. The fatty acid carbons contain one or more double bonds. The oxylipins are mainly derived from C16, C18, C20 or C22 PUFAs depending on the nature of PUFAs present in the algae. They can provide oil with the potential composition of saturated (palmitic (16:0), stearic (18:0)) and unsaturated (palmitolleic (16:1), oleic (18:1), linoleic (18:2), linolenic acid (18:3)). These compounds have unique compositions (higher levels of C18:3); in principle these compounds can be extracted and purified and can be used for different applications in pharmaceutical industry. The source of saturation and unsaturated lipids from algae can act as an alternative source of medically important molecules for useful drug formulations.

iii. Algal Lipid Extraction and Compositional Analysis:

Use of wet or dry biomass depends on the process being opted for biofuel production. Bligh and Dyer co-solvent method is the most commonly used method for extracting polar and nonpolar forms of lipids from algal biomass [Bligh, E.G. 1959]. Other techniques used for oil extraction are accelerated solvent extraction (ASE), super critical fluid extraction (SFE) and sub critical water extraction (SCWE) or hydrothermal liquefaction (HTL) [Pôjo, V.N.,2017]. Solvent extraction is most commonly used method but has some key limitations. Solvent extraction procedure by using soxhlet apparatus was used for extracting crude oil from dried algal-biomass (AOCS, 2003). Prior to extraction, dried algal cells were disrupted by employing sonication (5 min; 20 kHz) using n-hexane and taken into thimbles (30 g; made with Whattman Filter Paper No.1) for soxhlet extraction procedure. The mixture was refluxed for 5 h at 60 °C and concentrated in a rotavapor followed by vacuum drying with a temperature-controlled oil bath (120 °C) then cooled to room temperature. Thin layer chromatography (TLC) was performed to quantify the microalgal oil by using silica-gel as stationary phase, n-hexane and ethyl acetate (EA) (90:10) as mobile phase. Presence of fatty acids (FA), monoacylglycerides (MG), diacylglycerides (DG) and triacylglycerides (TG) in the microalgal oil was confirmed based on the mobility of the compound by comparing with pure vegetable oil.

Soxhlet extract was derivatized/transesterified to convert FA, MG, DG and TG to fatty acid methyl esters (FAMEs) by acid-catalysed methylation (Christie, 1982). In the one step transesterification procedure, the extracted lipid fraction along with 75 mL of 2% H_2SO_4 in methanol was refluxed for 4 h. After complete conversion (monitored by TLC), the solvent was partially removed and extracted with EA (2 × 100 mL). The combined EA layers were washed with distilled water (DW) to get the pH to neutral, dried over anhydrous sodium sulphate and evaporated under reduced pressure on a rotary evaporator.

iv. Fatty Acid Methyl Esters (FAME) Analysis

FAME composition was estimated by gas chromatography (GC; Agilent 7890B) using flame ionization detector (FID), capillary column (HP-88) and nitrogen as carrier gas. The temperature of the oven was initially maintained at 100 °C for 5 min, later increased to 250 °C at a ramp of 4 °C/min for 15 min. The injector and detector temperatures were maintained at 250 °C respectively with a split ratio of 50:1. FAME composition obtained was compared with the Standard FAME mix 37 component (Supelco).

v. Composition of Algal Oil

Based on the FID-GC analysis algal oil rich in both saturated fats (SF) and unsaturated fatty acids (USFA). SFAs and USFAs are long chain carboxylic acids with 12–24 carbon atoms. SFAs are saturated with hydrogen atoms without double bonds and possess economic importance such as combustion property, antiviral and antibacterial activity along with value- added applications in foods, detergents and cosmetics than USFAs. Whereas, USFA possess good lubricant property. C16:0 (Biofuel, and cosmetics preparation) which is having fuel property was found to be predominant and higher in concentration than other C:20, C11:0 (Biofuel, antiviral and antibacterial activity) and C14:0 (Biofuel, cosmetics and tropical medicinal preparation, C18:2 (cardiovascular diseases) C18:3 (antiviral), C22:0 (Good lubricant properties) and C22:2 (Good lubricant properties) components. Presence of C18:3 component in the algal lipid is good indication that algal oil can be used in pharmaceutical industry.

3.3 Functional Role and Mechanism of Linoleic Acid

Incubation of vesicular stomatitis virus with linoleic acid (a type of unsaturated fatty acid) can lead to leakage of viral envelopes. This effect was far more pronounced with a higher linoleic acid concentration, causing disintegration of viral particles (Figure 2).

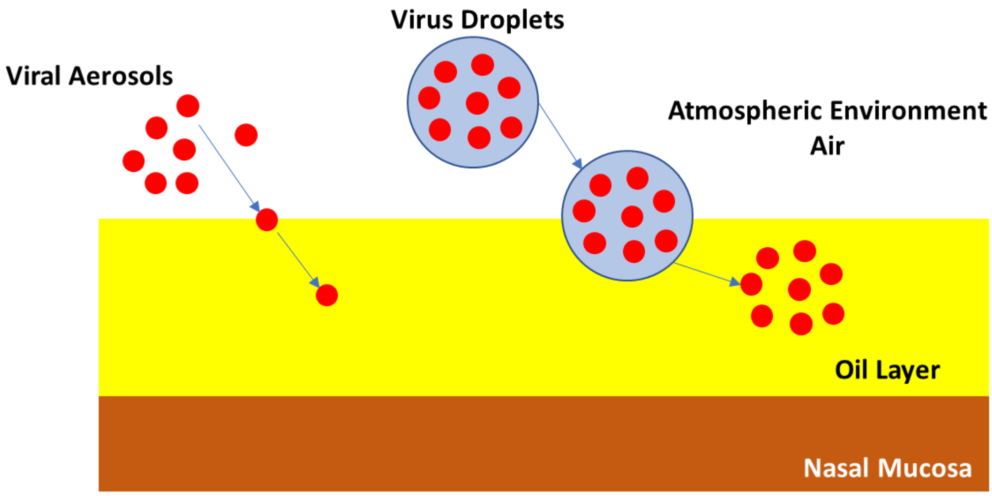

Fig 2: Possible adsorption process of virus-containing aerosols and droplets in air and oil interface

It was suggested that the antiviral mechanism is due to the fatty acids can be incorporated into the lipid membrane of viral envelopes, causing destabilization of the bilayer of viral envelopes [Thormar, Halldor, et al. 1987]. Interestingly, a recent study revealed that exogenous supplement of linoleic acid or arachidonic acid in human coronavirus 229E-infected cells was capable of significantly suppressing the coronavirus replication. This inhibitory effect was also observed in the highly virulent Middle East respiratory syndrome coronavirus [Yan, Bingpeng, et al.2019]. Thus, if the structures and functions of the COVID 19 S-protein or the viral envelope could be disrupted by sesame oil, the COVID 19 would be considered harmless to human body [Venkata Subhash et al., 2017; Kumari et al., 2013].

3.4 Overview on Using of Different Oils as Medicine

Ancient days the people have used different sources of vegetable sources of oils for body applications and mostly belongs to the Canola (rapeseed) oil, Soybean oil, Pumpkin seed oil, Perilla seed oil, Tofu and Walnut oil.

From the list sesame oil is attractive from historical experience because of its medicinal applications in treating of bacteria and virus. First, the oils are edible vegetable oil and derived from seeds. In the present manuscript ancient medicinal oil from sesame seeds is considered as example and used for comparative evaluation with algal oil. Chemical property of pure sesame oil has low surface tension and is incompatible with water and this is because of low intermolecular attraction between adjacent sesame oil molecules [Siddiqui, Nazima, and Adeel Ahmad., 2013]. For example, when the pure sesame oil was dropped in to water, it spreads out rapidly on the surface of water. So that when the sesame oil is coated to nostrils it may wet the nasal mucosa and act as a large protective layer and remain for a long time (boiling point of about 215 °C). Adhesion and thickness activity of protective oil layer in the nasal mucosa catch and trap the virus particles when a person is breathing and preventing the direct contact and binding of virus. Generally possible interface behaviour of virus-containing aerosols or virus-containing aqueous droplets at the air/sesame oil interface will be determined by the combined contributions of various effects (hydrophilic and hydrophobic properties of particles, particle size, air/oil surface tension, surface morphology, oil viscosity and oil density, etc) [Binks, Bernard P., and Tommy S., 2006]. The coating of sesame oil onto mucosal surface may increase the surface hydrophobicity of mucosal surface, and thereby decreasing the binding ability COVID 19 S-protein to mucosal cells.

Functional property like surface tension that of 75% ethanol solution the sesame oil behaves as same and may help to disrupt the surface structures and functions of the virus [Vazquez, Gonzalo.,etal., 1995]. Moreover, sesame oil is composed of various saturated or unsaturated fatty acids, including linoleic acid, oleic acid, palmitic acid and stearic acid, especially the unsaturated fatty acids, have been proven to be active against some enveloped viruses [Kapadia, Govind J., et al.2002; Thormar, Halldor, et al. 1987].

4 Conclusion

Preparation of oil based nasal sprays has potential to prevent the spread of virus from person to person by forming protective layer in the nostrils. As literature suggests, the oil nasal spray (main component linolenic acid) method involved in the inactivation of coronavirus to help spread of COVID 19. Apart from vegetable oil, the purified oil from algal source could be considered as a future option for such sprays and other formulations. We believe oil based nasal spray could help to prevent spread of coronavirus to fight COVID-19: Such nasal sprays could be especially useful for the asymptomatic infected population to inactivate coronaviruses in the upper respiratory tracts Application of oil could act as a surfactant or protective material also for inanimate objects such as surfaces of tables, door handles, our hands and faces for controlling the viral spread Use of oil sprays on napkin could be another possibility to cover nose and mouth before using it as a facial mask. The discussion in this paper provides some possible mechanisms that support use of purified algal oil to prevent infectious diseases such as COVID 19, However, proposed algal based oil nasal sprays should not be used until it has been verified and approved by future experimental and clinical studies and approved by the qualified government authority.

5 Declarations

5.1 Acknowledgments

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5.2 Authors Contribution

All authors equally contributed in the work which is reported in the present manuscript. Before publication of this manuscript, all the authors sincerely agreed with the terms and conditions of *AIJR Preprints*.

5.3 Competing Interests

The authors declared that there is no conflict of interest exist in the publication.

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