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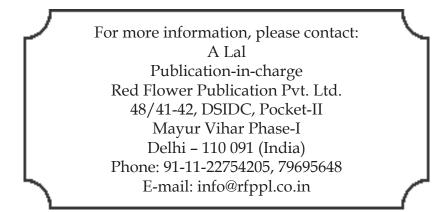
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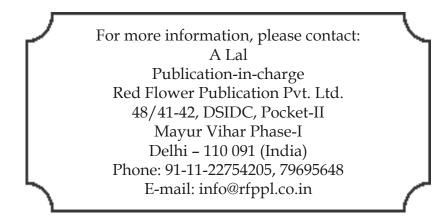


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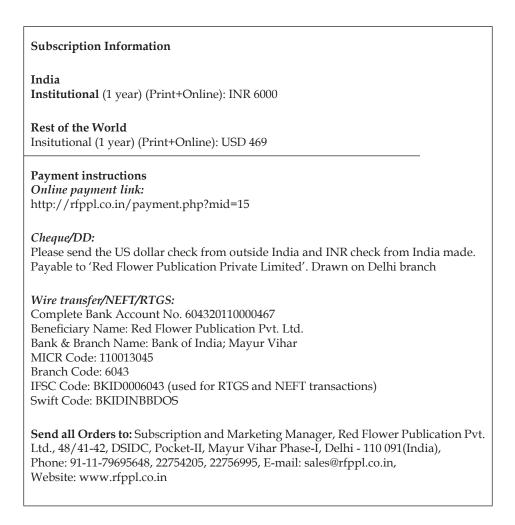
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Contents

Original Articles	
Risk Factors of Coronary Heart Disease (CHD): A Study on Knowledge Level of Population in Jorhat District of Assam Mridusmita Borthakur, Ruma Bhattacharyya, Sanjoy Borthakur	117
Review Articles	
Importance of Ayurveda for Management of COVID 19 Sangeeta Ahuja	123
Biofilm Inhibitory Potential of Chitosan based Nano-Encapsulated Phytochemicals: An Improved Antibiofilm Drug Delivery System for Antimicrobial Therapy Harshit Saxena, Viabhav Kumar Upadhayay, Meena Goswami, Vikas Pathak S K Bharti	129
Letter to Editor	
Post COVID 19 Patients: Chronic Fatigue Syndrome and Rejuvenation Through Nutrients Supplementation Neelesh Kumar Maurya	137
Subject Index	139
Author Index	140
Guidelines for Authors	141

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Risk Factors of Coronary Heart Disease (CHD): A Study on Knowledge Level of Population in Jorhat District of Assam

Mridusmita Borthakur¹, Ruma Bhattacharyya², Sanjoy Borthakur³

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Abstract

Coronary heart disease (CHD) is the most common type of heart disease and cause of heart attack. The risk of coronary heart disease increases as age increases. Middle aged adults are mostly affected by CHD. Although coronary heart disease incidences is increasing, it can be highly preventable as it is caused by many modifiable risk factors like smoking, alcohol consumption, obesity, physical inactivity, unhealthy diet, stress, high blood pressure etc. For prevention and reduction of the burden of coronary heart disease it is essential that people have knowledge of risks of Coronary heart disease so that they assume themselves at risk and adopt measures to prevent the disease. In the present study an attempt has been made to find out the knowledge level of population of Jorhat district of Assam towards risk factors of CHD. From the study it is observed that irrespective of sex both male (34.70%) and female (35.06%) maximum percentage of population had low level of knowledge regarding general nutrition. Majority of male (32.09%) and female (33.76%) respondents had low level overall knowledge about the risk factors. It is also recorded that the irrespective of sex the respondents had low level of existing knowledge regarding risk factors viz., diabetes, obesity and diet and risk factors of CHD. While in respect of hypertension, majority of respondents both male (38.06%) and female (36.53%) had medium level of existing knowledge. The incidence of CHD is likely to increase further because of rapid urbanization and its accompanying lifestyle changes. Therefore, there is an immediate need to raise awareness among the general population about these risk factors, promote the correct diet and physical activity.

Keywords: Coronary heart disease (CHD); Risk factors; Knowledge level.

Introduction

Coronary heart disease (CHD) is the most common type of heart disease and cause of heart attack. Cardiovascular disease (CVD), including coronary heart disease (CHD) and stroke, is the largest cause of mortality in the world, and the majority of deaths occur in low- and middle-income countries such as India and China. While the prevalence of CHD has

E-mail: mridubs@gmail.com

stabilized in developed countries, the condition has recently begun to impact developing countries due to increasing life expectancy, urbanization and lifestyle changes are become epidemic in urban locations of these countries and are rapidly increasing in rural areas as well (Fuster et al., 2010). In India, CVD is the largest cause of mortality in all regions of the country. The main approach to reducing the social and economic burden due to coronary heart disease (CHD) and other noncommunicable diseases is based on the identification of risk factors at the level of the individual (Nieto, 1999). This work began with the Framingham study in 1948 (Kannel, 2000) and has continued unabated. The risk of coronary heart disease increases as age increases. Middle aged adults are mostly affected by CHD. For men, the risk starts to climb at about age 45, and by age 55, the risk becomes double. It continues to increase until, by age 85. For women, the risk of coronary heart disease also climbs with

Author Affiliation: ^{1,4}Subject Matter Specialist, Department of Food and Nutrition, Krishi Vigyan Kendra, Assam Agricultural University, Golaghat 785619, India. ²Professor, Department of Food and Nutrition, Faculty of Community Science, Assam Agricultural University, Jorhat 785013, India. ³Senior Scientist and Head, Krishi Vigyan Kendra, Assam Agricultural University, Tinsukia 786126, India.

Corresponding Author: Mridusmita Borthakur, Subject Matter Specialist, Krishi Vigyan Kendra, Assam Agricultural University, Golaghat 785619, India.

age, but the trend begins about 10 years later than in men and especially with the onset of menopause (WHO, 2010). WHO has drawn attention to the fact that coronary heart disease is our "modern epidemic" a disease that affects populations, not an unavoidable attribute of ageing (Park, 2013).

Although coronary heart disease incidences is increasing, it can be highly preventable as it is caused by many modifiable risk factors like smoking, alcohol consumption, obesity, physical inactivity, unhealthy diet, stress, high blood pressure etc. Therefore, it is important that people know about the measures to prevent coronary heart disease by adopting healthy behaviours. For prevention and reduction of the burden of coronary heart disease it is essential that people have knowledge of risks of Coronary heart disease so that they assume themselves at risk and adopt measures to prevent the disease.

Thus, the baseline information about knowledge of coronary heart disease and its risk factors among the middle aged adults and identify the gaps in knowledge level as the modification in behaviour will not take place without adequate awareness of the health problem. In the present study an attempt has been made to find out the knowledge level of population of Jorhat district of Assam towards risk factors of Coronary heart disease (CHD).

Methodology

In order to study the knowledge level towards risk factors of Coronary heart disease (CHD) a survey has been conducted using a structured questionnaire with knowledge scale. A total of 810 respondents belonging to various socio-cultural organization i.e. clubs (161), societies (284) and samities (405) of Jorhat district were selected for the study. The statements used in the questionnaire were put into three point categories namely 'correct', 'incorrect', and 'do not know', with the scores 2, 0 and 1 respectively. Prior to data collection the knowledge scale was pre-tested on 10 samples. The data of the pre test were coded according to the order of the knowledge scale. All total 28 statements were used for final test battery after due assessment through a panel of expert. Thus the maximum possible total score varies from 0-56.

Results and Discussion

All total 810 respondents were surveyed with the knowledge scale, out of which 542 respondents

were female and 268 were male respondents. The socio-economic profiles of the respondents are presented in Table 1. From the Table it is observed that majority of the respondents were belonged to the age of 25–35 years.

Table indicates that 29.48 per cent of male respondents studied up to higher secondary level and 25.75 and 25.37 per cent of male respondents were HSLC passed and graduate while 2.24 per cent were post graduate. In case of female 32.84 and 30.26 per cent were under matric and HSLC passed, 18.46 per cent of respondents studied upto higher secondary level. Ali et al., 2016 reported that education level was also found to have a significant association with CHD knowledge.

More than 50 per cent of the male respondents were involved in service and followed by other profession like business and teaching. In case female 22.7 per cent of the respondents were involved in service and majority were unemployed.

It is evident from the Table that majority (79.85) of the male respondents were sedentary worker. In case of female more than 85 per cent of respondents were engaged in sedentary work while only 13.29 per cent of the respondents were moderate worker.

Table 1: Socio economic profile of respondents.

Sl. No.		Particulars	Male (N=268)		Fem	ale (N=542)
			No	Percentage	No	Percentage
1.		Age				
	a)	25-35	110	41.04	195	35.98
	b)	35-45	83	30.97	126	23.25
	c)	45-55	44	16.45	127	23.43
	d)	55-65	31	11.57	94	17.34
2.		Education				
	a)	Under matric	46	17.16	178	32.84
	b)	HSLC passed	69	25.75	164	30.26
	c)	HS passed	79	29.48	100	18.46
	d)	Graduate	68	25.37	87	16.05
	e)	Post graduate	6	2.24	12	2.21
	f)	Ph.D./ Doctorate	-	-	1	0.18
3.		Occupation				
	a)	Service	139	51.87	123	22.70
	b)	Business	94	35.05	24	4.43
	c)	Teaching	19	7.09	38	7.01
	d)	Unemployed	16	5.97	357	65.86
4.		Type of work				
	a)	Sedentary	-	79.85	-	86.71
	b)	Moderate	-	20.15	-	13.29
	c)	Heavy	-	-		

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Table 2: Percentage distribution of respondents according to presence of specific disease.

SL No.	Disease	Number	Percentage
1	Coronary heart disease	32	3.97
2	Diabetes	43	5.30
3	Hypertension	61	7.53

The Table 2 reveals that 3.97 per cent of respondents were suffering from coronary heart disease, 5.30 per cent of respondent were diabetics and 7.53 per cent of respondents were suffering from hypertension.

Existing knowledge of the respondents on risk factors of CHD

The existing knowledge of the respondents regarding general nutrition, hypertension, obesity, diabetes, diets and other risk factors of CHD like smoking, alcohol consumption, sedentary lifestyle and stress etc. are assessed and discussed in the following paragraphs.

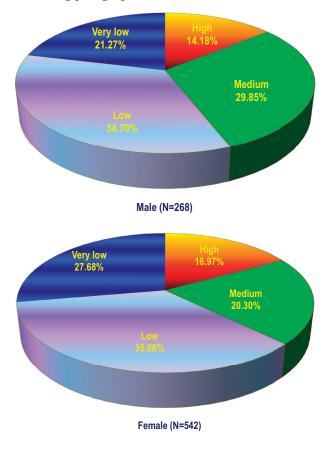


Fig. 1: Sex-wise percentage distribution of respondents according to existing knowledge level regarding general nutrition.

The Fig. 1 shows the sex-wise percentage distribution of respondents according to their

existing knowledge level regarding general nutrition. The Figure reveals that maximum percentage of male respondents (34.70%) and female respondent (35.06%) had low level of knowledge while 14.18 per cent of male respondents had high level of knowledge 16.97 per cent of female respondents had high knowledge level regarding general nutrition.

The knowledge level regarding different risk factors of coronary heart disease (CHD) is presented in the Table 3. The Table reveals that maximum percentage of male respondents (38.06%) had medium level of knowledge followed by 26.87 per cent low level of knowledge, 11.94 per cent very low level of knowledge, while 23.13 per cent of the male respondents had high level of knowledge in respect of hypertension. While, in case of female, 36.53 per cent had medium level of knowledge followed by 27.12 and 21.77 per cent of respondents had low and very low level, while 14.58 per cent of respondents had high level of knowledge.

Table 3: Existing knowledge level of the respondents regarding hypertension, obesity, diabetes, diet and other risk factors of CHD.

Male:	268
Female:	542

Sl. No.	Risk factors	Category	Knowledge level (%)			
			Very low	Low	Medium	High
1	Hypertension	Male	11.94	26.87	38.06	23.13
		Female	21.77	27.12	36.53	14.58
2	Diabetes	Male	23.13	29.85	25.75	21.27
		Female	18.08	39.85	22.32	19.74
3	Obesity	Male	17.54	41.42	17.54	14.92
		Female	23.80	36.72	18.82	20.66
4	Diet and other	Male	29.48	27.24	25.75	20.52
risk fa CHD	risk factors of CHD	Female	25.83	29.89	28.23	16.05
5	Overall existing knowledge level	Male	22.39	32.09	26.86	18.66
		Female	23.43	33.76	25.28	17.53

The results indicate that in respect of diabetes 29.85 per cent of the male respondents had low level of knowledge followed by 25.75 per cent with medium level. On the other hand in case of female, 39.85 per cent had low level of knowledge, whereas 19.74 per cent of the female respondents were found to have high level of knowledge regarding diabetes. Faisal et al., 2018 also highlighted the lack of knowledge and awareness of diabetes mellitus among the Saudi population.

WHO (2020) emphasizes that obesity is strongly related to major cardiovascular risk factors, such as

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raised blood pressure, glucose intolerance, type 2 diabetes, and dyslipidemia. The Table reveals that majority of the male respondents (41.42%) had low level of knowledge, while 26.12, 17.54, 14.92 per cent of the respondent had very low level, medium level and high level of knowledge respectively regarding obesity. In case of female maximum percentage (36.72) had low level of knowledge whereas 23.80, 18.82 and 20.66 per cent of the respondent had very low, medium and high level of knowledge respectively. Obesity is a key risk factor in the natural history of chronic non-communicable diseases such as diabetes mellitus, coronary heart disease and hypertension. Devi et al. (2001) stated that knowledge and associated opinions are responsible for either positive or negative attitudes towards obesity related aspects. Study conducted by Ali et al. 2016 stated that majority of the participants knew that being overweight increased the risk of CHD, fewer realized that carrying higher levels of abdominal fat imposes the greatest risk.

Diet is a major risk factor of coronary heart disease; other factors like high alcohol consumption accompanied by sedentary lifestyle and stress susceptible the younger adults towards coronary heart disease. The results indicate that majority of the male respondents (29.48%) had very low level of knowledge while 20.52 per cent had high level of knowledge regarding diet and other risk factors of CHD. On the other hand, almost equal per cent (29.89 and 28.23) of female respondents had low and medium level of knowledge, and 16.05 per cent of female respondents had high level of knowledge regarding diet and other risk factor like alcohol consumption, sedentary lifestyle and stress. Awad and Al-Nafisi, 2014 also reported sufficient knowledge levels among their Kuwaiti cohort regarding the risks associated with an unhealthy diet, smoking, obesity and physical inactivity.

It is evident from the Table that overall knowledge level of male respondents were slightly better than the female. Majority of female respondent (33.76%) had low level while maximum percentage of male respondents (32.09%) also had low level of knowledge, 26.86 per cent of male respondents had medium level of knowledge whereas 25.28 per cent of female had medium level, 18.66 per cent of male had high level while 17.53 per cent of female respondents had high level of overall existing knowledge.

Fig. 2 represents the percentage distribution of total respondents according to their overall existing knowledge level, indicates that majority of the respondents (32.84%) had low level of existing

knowledge, 26.05 per cent had medium level, 23.09 per cent had very low and 18.02 per cent had high level of existing knowledge. indicated an inadequate level of CHD risk factor knowledge among adult Omanis. Studies from other parts of the world have similarly reported low levels of knowledge and awareness of CHD risk factors (Haidinger et al., 2012 and Reiner et al., 2010). A study conducted in Kuwait to assess public knowledge of cardiovascular disease (CVD) risk factors reported similar findings, while another conducted in Jordan reported higher levels of knowledge among their cohort. (Mukattash et al., 2012; Awad and Al-Nafisi, 2014).

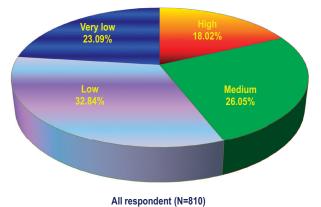


Fig. 2: Percentage distribution of total respondents according to overall existing knowledge level.

Conclusion

Coronary heart disease (CHD) is the most common type of heart disease and incidence of it is increasing, it can be highly preventable as it is caused by many modifiable risk factors like smoking, alcohol consumption, obesity, physical inactivity, unhealthy diet, stress, high blood pressure etc. For prevention and reduction of the burden of coronary heart disease it is essential that people have knowledge of risks of Coronary heart disease so that they assume themselves at risk and adopt measures to prevent the disease. From the present study it has been majority of the respondents had low level overall knowledge about the risk factors. The incidence of CHD is likely to increase further because of rapid urbanisation and its accompanying lifestyle changes. Therefore, there is an immediate need to raise awareness among the general population about these risk factors, promote the correct diet and physical activity, and at the same time develop guidelines for screening and preventive therapeutic programmes to identify and manage individuals at high risk for future CHD.

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Importance of Ayurveda for Management of COVID 19

Sangeeta Ahuja

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Abstract

Ayurveda is an ancient plant science. According to the type of the human body the combinations of doshas must be balanced. When the doshas become unbalanced the body showing some form of disease and disorder. The symptoms may be same for so many problems. So, the Ayurveda concentrate on the balancing the body that the doshas instead of something else but it totally depends upon the type of body. After identifying the body types the management of all diseases even Covid 19 can also manage to the large extent.

Data have been obtained and rigorous experimentation with statistical testing with inference have been done. The utilization of Ayurveda in life for balancing the doshas and increasing the immunity manage the treatment of patients suffered from Covid 19 gives promising results.

Keywords: Covid 19; Vata; Pitta; Kapha; Mind; Soul; Digestion; Meditation; Management; Happiness; Treatment; Spices; Herbs.

Introduction

Ayurveda is known as healing science or the "science of life". Ayurveda knowledge originated in India more than 5000 years ago. Ayurveda stems from the ancient Vedic culture. Many of natural healing systems have roots in Ayurveda. Ayurveda a Sanskrit word refers to the 'science of life and longevity'. Ayurveda adopts a holistic approach towards healthy living and is considered to be one of the oldest medicinal systems in the world. The base of Ayurveda philosophy to balance the body, mind and spirit. Both prevention and healing are carried out through natural means.

According to Ayurveda, each person is born with a life force that comprises the five elements or building blocks of nature earth, air, water, fire and space. We possess a unique balance of these five elements is known as Doshas. There are three fundamental dosha vata, pitta and kapha and a

E-mail: reach2san@yahoo.com

good health is considered to be a perfect state of balance between these three doshas.

The anatomical and physiological components of the human body are these three humors or doshas. They acts as constituents and protective barriers of the body in its normal physiological condition when out of balance. They contribute to disease process. The food and the individual actions nourish the humors and based upon the predominance of a particular humor, the human body is constituted. Accordingly each person is classified as vata, pitta and kapha.

For assessment of prikriti many factors have to be taken into consideration such as body build, skin, body temperature, hair, forehead, eyes, nose, lips, teeth, tongue, shoulder, joint, palms, nails, gait, muscles, physical activity, mental disposition, memory, speech and voice, sleep, sensitivity to weather, thirst, sweating, hunger, sweating, hunger, capacity of ingest food, food habits, capacity to digest food, Urine, Stool, Anger, Intolerance, Problem facing.

Ayurveda encourages the maintenance of health by maintaining the balance in one's life, right thinking, diet, lifestyle and the use of herbs. Each person is having unique pattern of energy i.e. physical, mental and emotional characteristics

Author Affiliation: Scientist (SS), Indian Agricultural Statistics Research Institute (ICAR), Ministry of Agriculture and Farmers Welfare, New Delhi 110001, India.

Corresponding Author: Sangeeta Ahuja, Scientist (SS), Indian Agricultural Statistics Research Institute (ICAR), Ministry of Agriculture and Farmers Welfare, New Delhi 110001, India.

which comprises their own constitution. The constitution is determined at conception by number of factors and remains the same throughout life.

Many internal and external factors acts upon us to disturb this balance and are reflected as a change in one's constitution from the balanced state. The solution is to understand the factors to nullify the cause of imbalance and reestablish the original constitution.

Through one's life, these constitutions may change based on physical, mental and emotional conditions. It is important to identify one's dosha and then create a lifestyle that suits it. This means adapting a diet and daily routine board on one's dosha. Ayurveda divided the different body into seven categories. Here it is possible for a person to be vata, pitta, kapha, vata-pitta, pitta-kapha, vatakapha or tri-dosha. However there is no best or perfect body types.

Everyone has a different combination of them and this determines one's genetically inherited physical and personality traits. Through one's life, these constitutions may change based on physical, mental and emotional conditions.

Various characteristics determine the dosha of an individual viz., body frame, body weight, skin type, eyes, hair, complexion, disease tendency, personality weakness, physiological traits, miscellaneous traits etc.

Energy is required to create movement so that fluids and nutrients get to the cells, enabling the body to function. Energy is also required to metabolize the nutrients in the cells and is called to lubricate and maintain the structure of the cell.

Vata is the energy of movement pitta is the energy of digestion or metabolism. Kapha is the energy of lubrication and structure. All people have qualities of vata pitta and kapha but the constitution is different. According to Ayurveda, the cause of disease is viewed as a lack of cellular function due to deficiency of vata, pitta or kapha. Disease can also be caused by the presence of toxins. For maintaining the balance in the body, mind and consciousness the understanding of vata, pitta and kapha is most.⁶⁻⁹

Vata, pitta and kapha are the combinations and permutations of space, air, fire, water, earth elements that manifest as pattern present in all creations. In the physical body vata is the subtle energy of movement, pitta is the energy of digestion and metabolism and kapha the energy that forms the body's structure. Vata is the subtle energy associated with movement composed of space and air. It governs breathing, blinking, muscle and tissue movement, pulsation of heart and all movements in the cytoplasm and cell membranes. In balance vata promotes creativity and flexibility. Out of balance, vata produces fear and anxiety. Pitta expresses as the body's metabolic system made up of fire and water. It governs digestion, absorption, assimilation, nutrition, metabolism and body temperature. In balance, pitta promotes understanding and intelligence. Out of balance, pitta arouses anger, hatred and jealousy. Kapha is the energy that forms the body structure-bones, muscles, tendons and provide the glue that holds the cells together formed from earth and water. Kapha supplies the water to all body parts and systems. It lubricates joints, moistures the skin and maintain immunity. Kapha is expressed as love, calmness and forgiveness out of balance, it leads to attachment greed and envy. To maintain the balance and health, diet and life style according to individual constitution strength the body, mind and soul. Drugs sometimes because of toxicity, weaken the body.

For balancing the kapha dosha one is suggested to be active, stay warm and active engage in stimulating activation avoid cold and dump things, cultivate physical challenges. For balancing the pitta dosha one has to be calm, rest and relax cut down striving avoid the sun. Stay cool. For balancing the vata dosha one has to be moderate adequate sleep disciplined schedule. Take in sun avoid over weakness stress, too much stimulation, wind and cold. To balance vata, pitta and kapha herbal remedies, warm oil massages, yoga and the diet is to be taken into consideration. To balance each dosha certain foods need to be consumed while some needs to be avoided.

One of the major reasons to kapha imbalance is excessive food consumption, and therefore a light, low fat diet of bitter, pungent and astringent tastes is recommended. This can include steamed or raw vegetables, ripe fruits, grains like oats, rye, barley and wallets, honey and strong spices like pepper, cardamom, cloves, mustard and turmeric. Kapha prikriti should avoid fats, milk and rice in their daily diet but can consume them occasionally.

High consumption of spicy and astringent foods attributes to vata imbalance. Hence a peace making diet with salty, sweet and sour tastes and warm, moist and easily digestable foods is recommended. This includes vegetables like broccoli, leafy vegetables and cauliflower, wheat and rice, mild spices like cumin, ginger and cinnamons. In addition, most foods like berries, melons and yogurt can help count-balance vata's as compared to other hydrating cooked foods such as soups or stews. Only foods like avocado, buttermilk, cheese, whole milk, coconut, nuts and seeds are supportive as well. People with vata prikriti should have slightly fatty foods and drink warm milk at night.

Excessive consumption of alcohol and spicy, oily, fried, salty, fermented food may result in pitta imbalance. They should avoid spicy, acidic or hot foods. Pitta balance can be restored with sweet, bitter and astringent tastes and heavy, cool foods such as sweet fruits, daily products, curry leaves, barley, oats and nuts. Avoid sour fruits potatoes, tomatoes, eggplant etc.

Irrespective of prikriti, one must always eat food on time, the ingredients should be seasonal and the meals should have a balance of all the ingredients required by the body to maintain good health and wellbeing.

Importance of Ayurveda in the Covid 19 pandemic

Most of the countries are facing the COVID 19 problem since last year. To deal with the impact of COVID 19 pandemic, Ayurveda plays a very important role.

Covid 19 epidemic disease various symptoms like body pain, cough, loss of taste, smell and abdominal pain, fever etc. Due to this deadly virus many people facing the hardships. Impact of lockdown completely effect the livelihoods earning of most of us. Workers are facing a lot 66% lost their jobs, 64% change in earnings, 77% households consuming less food than before, 47% household don't have enough money to buy even a week's worth of essentials., 77% vulnerable households received ration, 49% vulnerable households received a cash transfer.

Clinical trials using Ayurveda remedy for Covid 19 shows ground breaking results. Numerous countries and states have adopted or mandated natural treatment for novel coronavirus have shown mortality rates less because of utilizing the Ayurveda in life. It is matter of great pride for India that our ancient sciences are coming to the forefront with this new empirical evidence and sharing the instance of Ayurveda in the pandemic.

Experimental Analysis

Data have been obtained of patients having symptoms of Covid 19. The patients belongs to

different gender, age groups, profession. 5% of the total patients admitted having were Covid negative. Out of all Covid positive patients few have already suffered from some of the disorder/disease very few patients out of Covid positive were not having any disease in the past. All the corona positive patients have taken the ayurvedic treatment for the Covid recovery. Using the Advanced statistical technique Discriminate Analysis^{1-4,10,} prediction have been done. The recovery score have been computed. About 84% of the patients have been completely recovered within 22-25 days. In the first week, recovery was bit slowly, symptoms were very much visible. Gradually the observations were very much improved after utilizing the ayurvedic treatment. Out of 84%, 47% already had some disease/disorder in the past and 37% were not having any problem in the past (Fig. 2). Remaining 16%, 5% are Covid negative. Out of 11%, 7% are above age 69 to 78 years of age (male and female both) had low immunity and 4% are less than 45 years of age, were having low immunity.

There is no comparison of any type of treatment, the thing which is important is the recovery of the patients with less and no side effect. Since till now there is no vaccination of Covid. Ayurvedic treatment gives promising results. The test of significance have also been obtained at different level of significance.(Fig. 1 and 2)

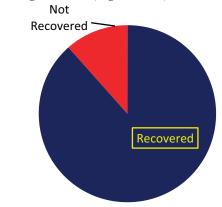


Fig. 1: Covid 19 Patients: Recovery.

Recovered

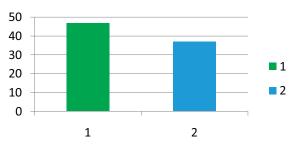


Fig. 2: Covid 19 Recovery with/without disease of history.

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Ayurvedic Treatment

Various things one has to keep in mind while management of Covid. Clean hands often, Cough and sneeze in the bent elbow, avoid touching one's eyes, nose and mouth. Limit social gatherings and time spent in crowded places, avoid close contact in some who is sick. Plant based science plays very important role. For uplifting and maintaining the immunity ministry of Ayush⁵ recommends the following self-care guidelines for preventive healing measures and boosting immunity.

Prevention is better than cure for happy and healthy life. It is true that stress and anxiety are part and parcels of a modern busy life, which cannot be ignored but it is essential to manage your stress level by practicing yoga or by other means, else in no time it can turn into a chronic heart condition.

Use boiled water instead of normal drinking water, use giloy, amla, herbal tea, cumin, moringa, tulsi, coriander, garlic, ginger, turmeric golden milk. Daily practice of yogaasans, pranayama and meditation for at least 30 minutes as advised by ministry of Ayush.⁵

For immunity promoting one can take chyawanprash 10 gm in the morning. Diabetic should take sugar free chanyanmanprash, Drink herbal tea/kadha made from basil, cinnamon, black pepper, dry ginger and raisin- once or twice a day.

Simple ayurvedic procedures (i) Nasal application Apply sesame oil coconut oil or ghee in both the nostrils in the morning and evening (ii) Oil pulling therapy Take 1 table spoon, coconut oil or sesame in the mouth and spit it off followed by warm water rinse. This can be done once or twice a day.

During dry cough/sore throat steam inhalation with fresh mint leaves or caraway seeds can be practiced once in a day. Clove powder mixed with natural sugar/honey can be taken 2–3 times a day in case of cough or throat irritation. These measures generally treat normal dry cough and sore throat.

The influence of mind and emotions on our immunity is a new system that is being studied in the past four/five decades known as psychoneuroimmunology. This studies the effect of the central nervous system on various factors responsible for immune response.

Many Factors that acts negatively on Immunity. The diet that is not nutritionally balanced and supportive to dosha balance. Excessed intake of processed sugars and additives for artificial flavoring is known to weaken the immune system. Excessive alcohol intake, irregular sleep, high stress, obesity, chronic medications, Dehydration.

There are so many factors that help augment immunity in Ayurveda. Aahra vihara nidra boost your immunity. A well balanced diet with all size tastes and qualities balanced based on ones dosha, Regular yoga practice, use variety of spices practice into the daily dietary routine. Most spices are high in their oxygen radical absorbance capacity and reduce cell damage and maintain integrity. Regular detox plants like fasting or proper panchkarma cleanse once a year. Consumption of rasayana herbs like ashwagandha and amla. Taking oil bath reduces stress levels and improve the quality of sleep which ultimately increase in immunity.

For getting peace of mind one should find some time for meditation which is a medium to get connectivity with GOD through the soul.

Conclusion

Ayurveda is an ancient plant science. Ayurveda is the balance of body, mind and soul. The body that is the prikriti of the individual is unique and the constitution of the body has been decided at the time of conception by the super power GOD. The constitution of the body are of one of seven kind basically a combination and permutation of vata, pitta and kapha. According to the type of the human body the combinations of doshas must be balanced. When the doshas become unbalanced the body showing some form of disease and disorder. The symptoms may be same for so many problems. So, the Ayurveda concentrate on the balancing the body that the doshas instead of something else but it totally depends upon the type of body. After identifying the body types the management of all diseases even Covid 19 can also manage to the large extent. The use of herbs, spices, hot water is must. The ratio doses are different for different body types.

The mind is related to neurons nervous systems which in terms of Ayurveda also have good/bad effect on health. To increase the energy happy hormones and optimism is very much needed. Soul is the interface between human and GOD. For peace of mind happiness yoga and meditation is must. For maintaining the body structural exercise is also needed. After balancing the body relaxing the mind and soul by maintaining the proper lifestyle, dietary habits and faith in GOD and oneself leads to perfect healthy wealthy happy life.

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Biofilm Inhibitory Potential of Chitosan based Nano-Encapsulated Phytochemicals: An Improved Antibiofilm Drug Delivery System for Antimicrobial Therapy

Harshit Saxena¹, Viabhav Kumar Upadhayay², Meena Goswami³, Vikas Pathak⁴, S K Bharti⁵

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Abstract

The production of biofilm by bacteria is the survival tactic in inappropriate environmental conditions. Biofilms also encourage pathogenic bacterial strains to develop antibiotic resistance. Exopolymeric substances (EPS) are the main constituent of biofilm that restricts the diffusion of plenty of antimicrobial compounds. Therefore, plant-derived compounds 'phytochemicals (flavonoids, tannins, alkaloids, terpenes, etc.) provide alternative options by showing the capability to inhibit biofilm produced by bacteria. However, the low stability, improper absorption, poor aqueous solubility, and easy degradability are few unfavorable features of phytochemicals that limit their use for further medical applications. However, the advancement in nanotechnology revolutionized antimicrobial chemotherapy, as various nanocarriers were shown their effectiveness in delivering bioactive compounds. Chitosan, derived from chitin, acts as bio carrier for a wide range of phytochemicals and improve their delivery and stability by exhibiting excellent antimicrobial and antibiofilm activities. The chitosan-based nanoparticles provided a feasible options to provide stability for encapsulated 'phytochemical/antibiofilm drug, and ensure its slow and proper release at the target site. Various methods such as ionic gelation, spray-drying' and 'emulsification' are used for encapsulating the natural bioactive components such as phytochemicals. The chitosan-based nanoparticles encapsulated with a number of phytochemicals namely ferulic acid, curcumin, cinnamaldehyde, chrysin, quercetin, and baicalein showed inhibitory potential against biofilm. This review presents a concise view of nano encapsulated phytochemicals for antibiofilm application, and development of efficient chitosan nanoparticles based antimicrobial therapeutic approach through effective drug (especially phytochemicals) delivery.

Keywords: Biofilm; Phytochemicals; Chitosan nanoparticles; Drug delivery.

Author Affiliation: ¹Student, College of Veterinary Science and Animal Husbandry, Uttar Pradesh Pandit Deen Dayal Upadhyaya Pashu Chikitsa Vigyan Vishwavidyalaya Evam Go Anusandhan Sansthan, Mathura 281001, Uttar Pradesh, ²Phd Scholar, Department of Microbiology, College of Basic Sciences and Humanities, Govind Ballabh Pant University of Agriculture and Technology, Pantnagar, Udham Singh Nagar, Uttarakhand, ^{3,5}Assistant Professor, ⁴Professor and Head, Department of Livestock Products Technology, College of Veterinary Science and Animal Husbandry, Uttar Pradesh Pandit Deen Dayal Upadhyaya Pashu Chikitsa Vigyan Vishwavidyalaya Evam Go Anusandhan Sansthan, Mathura (UP), India 281001.

Corresponding Author: Meena Goswami, Assistant Professor, Department of Livestock Products Technology, College of Veterinary Science and Animal Husbandry, Uttar Pradesh Pandit Deen Dayal Upadhyaya Pashu Chikitsa Vigyan Vishwavidyalaya Evam Go Anusandhan Sansthan, Mathura (UP), India 281001.

E-mail: dr.goswami2008@yahoo.co.in

Introduction

Biofilm formation is the unique trait in bacteria which assist their survival in different environmental settings. The clinical pathogens were also determined to possess the ability of biofilm formation and thereby engender a risk of antibiotic resistance (Høiby et. al., 2010). Extracellular polysaccharides are the main architectural component of the biofilm, and besides, biofilm also possesses extracellular proteins and DNA. These components serve as a blockade for the antimicrobial compounds given during the treatment (Khan et al., 2020a; Limoli et al., 2015). Therefore, a higher dose of the combination of antibiotics is administered to treat the microbial biofilm infection, but it also results in the development of antibiotic resistance pattern in bacteria and also induction of formation of recalcitrant biofilm (Khan et al., 2020b).

So, there is a need to explore novel antimicrobial compounds and the development of effective strategies to combat bacterial infections (Römling et al., 2012). 'Herbal plants', the eminent gift endowed by nature, are the hub of 'phytochemicals' possess a ample range of medicinal values (Gupta et al., 2017). Numerous phytochemicals was assayed for their antibiofilm trait against several clinical pathogens (Ta et al., 2015). But the low bioavailability, poor stability, and nonsoluble nature in aqueous solution limit the potential of phytochemicals for using as biofilm inhibitory agents (Akolade et al., 2017; Gopalakrishnan et al., 2014). So, the advancement in nanotechnology further illustrated the nanoencapsulation of phytochemicals or other natural bioactive components for further development of antimicrobial therapy (Mu et al., 2016).

The natural polymer especially chitosan is potentially used as a bio-carrier in order to get desirable effects of loaded phytochemicals. The chitosan-based nanoparticles also exhibited the antibiofilm effect as it can easily diffuse into the biofilm composite structure (Chávez de Paz et al., 2011). 'Ionic gelation', 'spray-drying', and 'emulsification' are the three key methods employed in the chitosan-based nanoencapsulation of an antibiofilm agent (phytochemicals, herbal extract, antimicrobial drug) (Akbari-Alavijeh et al., 2020; Detsi et al., 2020; Khan et al., 2020b). 'Chitosan nanoparticles', exhibited the excellent potential for the delivery of numerous phytochemicals and other drugs for inhibiting biofilm formed by bacterial pathogens (Ilk et al., 2016; Khan et al., 2020b). The main benefits of nanoencapsulation are to slow and constant release of the particular embedded phytochemical or drugs at the target location. Moreover, chitosan based nanoparticles present numerous benefits in terms of better effectiveness, economically feasible, eco-friendly, augmented absorptibility (Ilk et al., 2017; Pattnaik et al., 2018a).

Biofilm formation: an unique mechanism of pathogenic bacteria

The drug resistance mechanism in bacteria is well known, and the formation of biofilm is a major factor which engenders the pattern of antibiotic resistance in clinical pathogens (Høiby et al., 2010; Wu et al., 2015). Bacteria establish biofilm on biotic and as well as abiotic surfaces through secreting extracellular polymeric substances. The biofilm matrix, besides the exopolymeric substances, also contains other components including carbohydrate-binding proteins, extracellular DNA, pilli, adhesive fibers, and flagella (Kostakioti et al., 2013). The biofilm matrix contains some unique

features that provide protection to bacterial cells from numerous external environmental factors. The bacterial cells show tolerance or resistance pattern towards antibiotics and cells in biofilm may exhibit a thousand times antibiotic resistance ability than the 'planktonic cells' (Khan et al., 2020a). Formation of biofilm is the tactic adopted by bacteria for their survival in hostile conditions (Wu et al., 2015). There are the following main three stages for the development of biofilm on surfaces including a) attachment of microbial cells to an abiotic/biotic surface, b) aggregation of microbial cells to construct microcolonies, and finally, c) differentiation of biofilm into full-grown structural components (Roy et al., 2018).

In the clinical settings, biofilm-producing bacterial cells are usually the source of nosocomial, chronic, and medical/ implantable device-associated infections (Khatoon et al., 2018). Bacterial produced biofilms are coupled with human ailments and also responsible for a number of biofilm-associated infections such as chronic obstructive pulmonary diseases, lung infections of Cystic Fibrosis) patients, otitis media, chronic wound infections, rhinosinusitis, endocarditis, vaginosis, urinary tract infections, inflammatory bowel disease, prostatitis, cancer, etc. (van Tilburg Bernardes et al., 2015; Vestby et al., 2020). A number of clinical isolates such as 'Pseudomonas aeruginosa', 'Burkholderia cepacia', 'Listeria monocytogenes', 'Staphylococcus aureus' and 'Candida albicans', as causal agents of nosocomial infections in immune-compromised people were reported to form the biofilm structure (Ta et al., 2015).

Phytochemicals: as effective antibiofilm agents

'Phyto-therapy' employed the use of herbal plants for combating against several ailments. A number of parts such as leaves, flowers, seeds, root, rhizome, etc are used for remedial purposes in various ethnic communities (Kumar et al., 2017). Plants are the ultimate hub of numerous compounds commonly known as 'phytochemicals'. Particularly, the health benefit effects are ascribed due to the phytochemicals (Mohanraj et al., 2018). In the last few decades, the emergence of antibiotic resistance in pathogenic bacteria posing problems, and therefore there is a requisite to search for other approaches such as using phytochemicals for combating microbial infections (Gupta et al., 2017).

Biofilm formation by bacteria also decreases the effect of antibiotic and presents the major mechanism of antibiotic resistance. Therefore, phytochemicals from the herbal sources may provide an alternative

solution for inhibiting biofilm formation in bacteria. Various phytochemicals such as plant phenolics (including'benzoates', 'phenylpropanoids', 'stilbenes', 'flavonoids', 'gallotannins', 'proanthocyanidins' and 'coumarins'), terpenes, alkaloids, guinones, and organosulfur compounds were showed remarkable antibiofilm activity against clinical pathogens (Ta et. al., 2015). Eugenol, a 'phenylpropene' showed antibiofilm potential against one clinical pathogenic strain of Pseudomonas aeruginosa and reduced biofilm formation (43% at 400 µM) (Zhou et al., 2013). Tannins were also tested for their antibiofilm and quorum sensing inhibitory properties. 'Punicalagin' an example of tannin (at the concentration 15.6 µg/ml) was reported to downregulate the expression of motility and QS related genes in Salmonella typhimurium (Li et al., 2014).

Tannic acids were determined for biofilm inhibitory effect against Pseudomonas and Staphylococcus aureus (Ta et al., 2015). Flavonoids such a quercetin (at 1 μ g/ml concentration) inhibited biofilm production in MRSA (>80%) and MSSA (>50%) strains and also declined the expression of genes participated in QS and virulence of Staphylococcus aureus (10 µg/ mL) (Lee et al., 2013). Other flavonoids namely 'catechins' isolated Camellia sinensis L. also inhibited biofilm production in Eikenella corrodens (Matsunaga et al., 2010). Various kinds of terpenes (such as 'monoterpenes', 'limonoids', and 'triterpenes') were also been found to contain anti-biofilm and anti-quorum sensing activities (Ta et al., 2015). Monoterpenes namely thymol and carvacrol had shown efficacy against biofilms of Listeria monocytogenes (Upadhyay et al., 2013), while the 'sesquiterpenoids' (such as salvipisone acanthospermolides) reduced and biofilm formation in Pseudomonas aeruginosa (Cartagena et a., 2007). Moreover, allicin (sulfur containing phytochemical) inhibited biofilm formation in Staphylococcus epidermidis strains (at 4 mg/ml) (Pérez-Giraldo et al., 2003). At a concentration of $63.5 \,\mu\text{g/ml}$, 'berberine' (an example of alkaloids) reduced biofilm growth in different clinical isolates of Klebsiella pneumoniae (Magesh et al., 2013). Coumarins also exhibited QS and biofilm distorting activities, as 'aesculetin', at the level of 500 µM, was found to inhibit QS in C. violaceum, E. coli and P. aeruginosa (Brackman et al., 2009).

Chitosan-nanoparticles as effective antimicrobial drug delivery tool

Chitosan is an example of a linear polysaccharide comprised of 'D-glucosamine (GlcN)' and

'N-acetyl glucosamine (GlcNAc)' connected by β -1, 4-glycosidic bond. It is synthesized from the chitin (second huge polymer after cellulose, exist in the body of crustacean, mollusks, and insects) after the partial alkaline deacetylation (Jung et al., 2014). Either homogenous or heterogeneous deacetylation methodologies are applied for deriving the chitosan from the chitin (Aam et al., 2010). On the contrary, chitooligosaccharides and other derivatives of chitosan are produced either by enzymatic or acid hydrolysis from chitosan (Khan et al., 2020). Molecular weight (MW) and extent of deacetylation of chitosan and chitooligosaccharides are two key factors for determining their biological activities (Foster et al., 2015). Furthermore, chitosan and chitooligosaccharides can be used as 'biocarriers' for the phytochemicals or other natural compound showed antibiofilm activity.

As carrier molecule, the chitosan/ а chitooligosaccharides can improve the drug delivery and maintain the stability of encapsulated plant-derived compounds. Numerous studies exhibited limitations of antibiofilm drugs (including phytochemicals) such as poor stability, low bioavailability, degradation, and water insolubility (Dos Santos Ramos et al., 2018). In order to fight such challenges, the controlled release strategy is emerging where the antibiofilm compounds are loaded to the various preparations of chitosan as 'carrier molecules' (e.g. 'nanocomposites', 'microspheres', 'nanofibers', 'hydrogels', and nanoparticles) (Bilal et al., 2019). The benefits of such carrier molecules are to facilitate the controlled release of phytochemicals having antibiofilm potential such as quercetin (Omwenga et al., 2018), cinnamaldehyde (Pattnaik et al., 2018a), ferulic acid (Dasagrandhi et al., 2018), caffeic acid (Kim et al., 2018), and kaempferol (Ilk et al., 2016) which had resulted in extended effectiveness and the slightest cytotoxicity effects. Moreover, the natural antimicrobial agents such as phytochemicals, and essential oils encapsulated within chitosan nanoparticles were more stabilized. Encapsulated antibiofilm agents were also capable of protecting themselves from environmental degradation (Khan et al., 2020).

Need of phytochemical encapsulated chitosan nanoparticles

Using phytochemicals for their wide spectrum of pharmacological applications is widely accepted as they are effective and safe. But some inappropriate behaviors such as low bioavailability, degradation, Harshit Saxena, Viabhav Kumar Upadhayay, Meena Goswami et al. \ Biofilm Inhibitory Potential of Chitosan based Nano-Encapsulated Phytochemicals: An Improved Antibiofilm Drug Delivery System for Antimicrobial Therapy

poor stability, and adsorption limit the uses of phytochemicals (Gopalakrishnan et al., 2014). Often phytochemicals showed potential bio-activities under laboratory or in vitro conditions becomes fail in terms of activities and efficiency under in vivo studies and clinical trials (Akolade et al., 2017). Khan et al, (2014) illustrated the low absorption of phytochemical especially polyphenols after ingestion from the gastrointestinal tract. Other phytochemicals, for instance 'catechin', depicts unstable nature in gastrointestinal solutions (Dube et al., 2011). Some bioactive compounds experience the process of enzymatic oxidation and show the nature of degradability in plenty of food processes and storage and may form components bearing harmful effects (Detsi et al., 2020). Reduced aqueous solubility of numerous phytochemicals is a further drawback (Akolade et al., 2017; Detsi et al., 2020). Additionally, numerous agents (either the surfactants or solubilizing agents) used in the process of extraction of the bioactive components might reduce their concentration (Akolade et al., 2017). The augmented dosage of phytochemicals requisite for the preferred curative effectiveness is also posing a strong limitation of their utilization (Yadav et al., 2012). This frequently results in selective or functional toxicological complications because of loss, inactivation, and/or degradation at the period of transportation of the molecule from the site of administration to the target location (Akolade et al., 2017).

Therefore, it becomes imperative to seek a carrierbased system for the phytochemicals which could abolish most of abovementioned limitations with the aim of improving the clinical results (Khan et al., 2014). So, 'nanoencapsulation' is a proficient tactic to conquer these limits through following such as a) increasing the targetability, b) prolonging the rate of release of the encapsulated bioactive components and, c) improving the stability of the encapsulated material. After ingestion, nanoparticles containing 'phytochemicals' may possibly adhere to the mucosa of the gastrointestinal tract, because of their 'mucoadhesive traits' (Khan et al., 2014), and then be transported through the circulation to various organs-targets extending their remedial effects (Detsi et al., 2020). The Barros and Casey, (2020) advocated the use of nano encapsulated plant extracts, essential oils and isolated pure phytochemicals as they become more effective weapons against biofilm.

Preparation of phytochemical encapsulated chitosan nanoparticles

There are three important methods namely 'ionic gelation', 'spray-drying' and 'emulsification' used in the chitosan-based nanoencapsulation of natural products or phytochemicals (Akbari-Alavijeh et al., 2020). The method of ionic gelation is simple which presents an organic solvent-free strategy for the development of stable nanoparticles (Detsi et al., 2020). It is based on the interaction amid oppositely charged macromolecules and nonhazardous and multivalent material in order to provide the charge density. Ionic gelation results owing to cross-linking of the 'polycationic chitosan' (both inter and intra cross liking) via an anionic cross-linker such as tripolyphosphate. For this, the chitosan firstly dissolves in an aqueous solution of acetic acid followed by the dropwise addition of tripolyphosphate. The formation of nanoparticles occurs at room temperature (under mechanical stirring) (Desai et al., 2016; Detsi et al., 2020; Wang et al., 2016). The elevated loading ability is determined as the benefit; but, the huge particle sizes, pH sensitivity, and higher polydispersity are the major downsides of this technique (Das et al., 2019; Shetta et al., 2019).

The second approach is the spray drying method which is carried out in the following steps a) dissolving of chitosan in acetic acid solution to form the matrix, b) homogenization of compound to be encapsulated with matrix, c) addition of selected cross-linking agent such as tripolyphosphate (Oliveira et al., 2005) or d, l-glyceraldehyde (Ravi Kumar et al., 2000; Wang et al., 2016), e) atomizing mixture in a chamber by a nozzle or spinning wheel, f) evaporation of water by hot air contacting the atomized material, and collection of material fall to the bottom of the dryer (Detsi et al., 2020; Gibbs et al., 1999). This approach is considered to be simple, fast, and economically feasible (Detsi et al., 2020).

Furthermore, the third approach is illustrated as the 'emulsification-solvent diffusion technique'. This method is relied on the crosslinking of functional amines groups of 'chitosan' and 'aldehyde' (for instance 'glutaraldehyde', 'formaldehyde') or even 'vanillin' as an ecofriendly substitute. The precipitation of chitosan takes place upon the organic solvent diffusion into water (Detsi et al., 2020; Wang et al., 2016). Various phytochemicals such as flavonoids (e.g., genistein) (Rahmani et al., 2020), tea polyphenols (Dube et al., 2011; Liang et al., 2017), curcumin (Almutairi et al., 2020), baicalein (Ahmed et al., 2020), quercetin (Omwenga et al., 2018), kaempferol (Ilk et al., 2017), phenolic compounds such as ferulic acid (Panwar et al., 2016) were encapsulated in chitosan nanoparticles, and also determined tested for therapeutic approaches.

Antibiofilm effect of chitosan nanoparticles loaded with the phytochemicals

phytochemicals Nano-encapsulated were potentially determined for their antimicrobial activity against pathogenic bacteria (both Gramnegative and Gram-positive) and fungi. But very few reports are reported on using nano encapsulated photochemical as 'antibiofilm agents' where nanocarrier improved the delivery of phytochemicals for distorting bacterial produced biofilm. Two flavonoids, quercetin, and baicalein, in associated with chitosan-based nanocapsules exerted inhibiting potential against quorum sensing in bacteria and biofilm formation (Omwenga et al., 2018). 'Ferulic acid grafted chitosan nanoparticles (FA-CSNPs)' prepared by employing ionotropic gelation method exerted potential antibiofilm activity against Candida albicans (Panwar et al., 2016). The study of Jahanizadeh et al., (2017) showed the role of novel bio nanocomposite of 'Carboxymethyl Starch (CMS)-Chitosan (CS)-Montmorillonite (MMT)' for the delivery of curcumin, and illustrated the effective role of curcumin loaded bio-nanocomposite against biofilm on dental models formed by Streptococcus mutans.

Kaempferol encapsulated chitosan nanoparticles were found to inhibit quorum sensing (QR) related processes (as this encapsulated material inhibited the production of a pigment 'violacein' in bacterial strain namely Chromobacterium violaceum CV026) and thus provoked for developing antimicrobial therapy as stable quorum sensing (QS)-based antibiofilm agent (Ilk et al., 2017). 'Cinnamaldehyde encapsulated chitosan nanoparticles' was reported to down-regulate the quorum sensing linked virulence and biofilm production in Pseudomonas aeruginosa PAO1 (Pattnaik et al., 2018a). Moreover, Pattnaik et al, (2018b) exhibited the potential antibiofilm property of 'ferulic acid encapsulated chitosan-tripolyphosphate nanoparticles (FANPs)' against Pseudomonas aeruginosa in comparison of 'nonencapsulated ferulic acid'. The study also illustrated the promising features of nano encapsulated ferulic acid such as anti-quorum sensing activity and slow and constant release of

ferulic acid at the targeted location. Furthermore, Dasagrandhi et al., (2018) showed antibacterial and antibiofilm the activity of 'ferulic acid-grafted chitosan (CFA)' against Listeria monocytogenes, Pseudomonas aeruginosa, and Staphylococcus aureus. The antibiofilm ability of chitosan implanted with phenolics (such as caffeic acid, ferulic, and sinapic acid) were determined against foodborne pathogens such as Pseudomonas aeruginosa and Listeria monocytogenes (Kim et al., 2018). 'Chrysin-encapsulated chitosan nanoparticles (CCNPs)', prepared by emloying the ionic gelation methodology exhibited excellent anti-biofilm activity against Staphylococcus aureus (Siddhardha et al., 2020).

Conclusion

The nano-encapsulated phytochemicals are being used in current researches to explore their antimicrobial and antibiofilm activities. Using chitosan-based nanoparticles offers a better strategy to maintain the stability of embedded phytochemicals, and thereby show excellent antibiofilm activity through improvement in delivering phytochemicals at the target site. However, very few studies have been done for decoding antibiofilm activity in terms of chitosanbased nano encapsulated pure phytochemicals. More investigations are still required for encapsulating wide arrays of phytochemicals through chitosan-based nanoparticles in clinical trials.

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Neelesh Kumar Maurya

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A novel coronavirus was first identified in Wuhan, China, in December 2019. The World Health Organization named coronavirus 2 acute respiratory syndromes (SARS-CoV-2) and is responsible for coronavirus disease in 2019 (COVID-19).¹ 33,998,571 Globally, recorded cases of COVID-19, including 1035636 fatalities, were announced to the worldometers.info on 3 October 2020.² Present coronavirus disease (COVID-19) has been global health and economic pandemic. Infection with extreme acute respiratory syndrome-coronavirus-2 (SARS-CoV-2) is known to cause severe respiratory symptoms to acute respiratory distress syndrome (ARDS). The highest mortality rates have been reported in patients with underlying cardiovascular disease and elevated cardiac troponin levels. This high inflammatory load is believed to be co-responsible for ARDS growth, vascular inflammation, myocarditis, and other cardiac events.3 Scientists observed that patients who contracted COVID-19 displayed 'cytokine storm' patients had elevated levels of IL-2, IL-7, granulocyte-colony stimulating factor, and tumor necrosis factor. The virus is associated with delayed expression of type I interferon (IFN) signaling, a central part of the innate defense against viral infections. A previous study has demonstrated a complicated relationship between immunity and genetic expression where the infectious agent enhances proinflammatory cytokines, such as interleukin (IL)-1, IL-6, and tumor necrosis factor, with modifications in T-cells and natural killer cell function.⁴ Over expression of these immune modulators induces fatigue so that the host retains the resources required to battle

E-mail: neeleshkumar.maurya@gmail.com

the infection. An example of this is when we used viral hepatitis interferon. Few patients may have endured this medication due to impaired fatigue and stress-induced by interferon. The previous research indicates a history of hyper inflammation that was consistent with problems such as multiorgan failure.5

COVID-19 is not a short-term critical health care problem that would go down and not return. Recovered COVID-19 patients are incapable to perform normal routine task and feel lack of energy. Additional organ structures that may be impaired include the brain, liver, and kidneys. Depression, fear, and post-traumatic stress injury are also possible long-term sequels to COVID-19.6 The continuous process would comprise of an interminable number of patients with compromised functional status and quality of life who should be treated as super specialties physicians through health multidisciplinary teams. It should be remembered that chronic fatigue is a complicated condition, and no recovery plan works for everyone. Multi-faceted treatments are usually used to effectively control the syndrome's effects, including incapacitating exhaustion and discomfort. Studies indicated that these nutrient supplements help combat post-COVID 19 fatigue in patients.⁷

- Vitamin B12, folic acid as RDA
- Selenium as RDA
- Vitamin D: 10 mg per day
- Magnesium malate 1000 mg, 150 mg of El. Three a day.
- Zinc gluconate 15 to 30 mg per day
- Acetyl-L-carnitine 0.5 to 2 g twice daily (may be increased to 2 g three times daily if required in extreme cases)
- Fish oil (EPA / DHA) 2 g a day
- Co-Q 10, 100 to 200 mg per day

Author's Affiliation: 1Research Scholar, Department of Food and Nutrition, Institute of Home Science, Bundelkhand University, Jhansi, Uttar Pradesh 284128, India.

Corresponding Author: Neelesh Kumar Maurya, Research Scholar, Department of Food and Nutrition, Institute of Home Science, Bundelkhand University, Jhansi, Uttar Pradesh 284128, India.

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Subject Index

Page No

Biofilm Inhibitory Potential of Chitosan based Nano-Encapsulated Phytochemicals: An Improved Antibiofilm Drug Delivery System for Antimicrobial Therapy	129
Climate Change and Childhood Malnutrition	23
Dietary Fiber and Mineral Enriched Carrot Pomace Powder Bread	65
Importance of Ayurveda for Management of COVID 19	123
Knowledge, Attitude and Practice of Nutrition: Adolescents Study	15
Life Chakra Mantra for Work Life Balance	9
"Multi-Potential Influence of Gut Microbiota on Human Health"	79
Nutrimental and Ethnomedicinal Potential Plants of the Qur'an: An Overview - IV	29
Need of Milk Fortification and Indian Scenario	45
Nutrimental and Ethnomedicinal Potential Plants of the Qur'an: An overview: V	85
Post COVID 19 Patients: Chronic Fatigue Syndrome and Rejuvenation Through Nutrients Supplementation	137
Risk Factors of Coronary Heart Disease (CHD): A Study on Knowledge Level of Population in Jorhat District of Assam	117
To Study the Effect of Imparting Nutrition Education on Nutrient Intake of 10-11 Year old Girls of Government Schools in Mumbaiy	73
Trans: Fat in Modern Diet	19

Author Index

Name	Page No	Name	Page No
Barkha Sharma	51	Ruma Bhattacharyya	23
DY Athina Deepa Prasanna	19	R L Rajput	65
E Lakshmi	15	Rekha Battalwar	73
Harshit Saxena	129	Ruma Bhattacharyya	117
K Silambu Selvi	19	Sangeeta Ahuja	9
Kamaliya Keshav B	65	Sangeeta Ahuja	123
Meena Goswami	51	Sanjay Singh	51
Meena Goswami	129	Sanjoy Borthakur	117
M B Kapopara	65	S K Bharti	129
Megha Khattar	73	TP Mall	29
Mamata Mishra	79	T P Mall	85
Mridusmita Borthakur	117	Urmimala Baruah	23
Neelesh Kumar Maurya	137	Vivek Sahu	51
Priya	51	Vikas Pathak	51
Pratibha Yadav	51	Vikas Pathak	129
Prasanna Kumar Dixit	79	Viabhav Kumar Upadhayay	129
Patel Devesh H	65		

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Standard journal article

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[9] National Statistics Online – Trends in suicide by method in England and Wales, 1979–2001. www. statistics.gov.uk/downloads/theme_health/HSQ 20.pdf (accessed Jan 24, 2005): 7–18. Only verified references against the original documents should be cited. Authors are responsible for the accuracy and completeness of their references and for correct text citation. The number of reference should be kept limited to 20 in case of major communications and 10 for short communications.

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