

Evaluation of oxidative biomarkers in relation with the self-reported symptoms among extensive usage of smartphone users on the basis of Age

By

Shirley A

(20PBT014)

Supervisor

Dr. K.V Shalini

A thesis Submitted to

Avinashilingam Institute for Home Science and Higher Education for Women,
Coimbatore – 641043

In Partial Fulfilment of the Requirement for the Degree of

Masters of Science in Biotechnology

May 2022

Certificate

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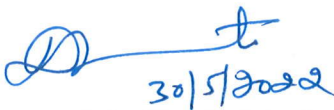
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Signature of the Head of the Department



Signature of the Supervisor

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Introduction

INTRODUCTION

Mobile phone is the most dominant portal of information and communication technology. In earlier days mobile phones are used for the communication purpose alone like to communicate with people who are far away. In the last few years, hand phones have become an integral part of our lives. (Parasuraman *et al.*, 2017). The mobile cellular subscriptions are constantly increasing every year (Peraman R, 2016). It is used for entertainment, social media, reading books, shopping, online business, gaming, study purpose, photography, videography and watching movies.

Over-usage of mobile phone may cause so many complications including eye irritation, wrist pain, eye dryness, weakness of thumb and wrist, neck pain, computer vision syndrome, insomnia, sleep disturbance, stress and hallucination (Peraman, 2016). In recent years mobile phone usage is increased in schools and colleges because of online classes due to pandemic situation. The signs of smartphone addiction are constantly checking the phone for no reason, feeling anxious or restless without the phone, waking up in the middle of night to check the mobile and communication updates, delay in professional performance as a result of prolonged phone activities, and distracted with smartphone applications (Chen, 2016). This leads to daytime tiredness, and reduced sleep duration.

The smartphone users tend to report pain in the neck, shoulder, and thumb, and the severity of the symptoms increases as the total time spent using the smartphone (Sang in Jung *et al.*, 2016). Musculoskeletal symptoms due to intensive texting on a mobile phone have been reported by Ming, *et al.*, (2010), and techniques used for text entering have been studied in connection with developing musculoskeletal symptoms (Gustafsson *et al.*, 2010). People tend to be in reflexed position for a longer period of time due to addiction. Such position can lead to neck pain and shoulder pain. Younger people are likely to use mobile phone during bedtime. They do chat, play games, watch videos, scroll social media without concerning about the time. It is reported that if mobile phone is used when light is off it transmit blue light to our eyes which would cause myopia, eye irritation, eye problems, redness etc.

Studies have highlighted associations of smartphone addiction with reduced productivity and with lower academic attainment, demonstrating the negative functional impact on young people's lives and future prospects. Indirect harms resulting from smartphone

addiction include propensity for accidents, for example through use whilst driving, and potential contribution to the obesity crisis by facilitating sedentary lifestyles (Taneja., 2005). Although at an early stage, there is also some neuroimaging evidence of volume and activity parallels between smartphone addiction and other addictions. A mental impairment resulting from modern technology has come to the attention of sociologists, psychologists, and scholars of education on mobile addiction. Mobile phone addiction and withdrawal from mobile network may increase anger, tension, depression, irritability, and restlessness which may alter the physiological behaviour and reduce work efficacy (Babadi-Akashe *et al.*, 2015)

Not only integral problems research has shown an increased risk of traffic accidents, about 3-4 times greater chance of an accident, when mobile phones (either handheld or with a "hands-free" kit) are used while driving due to distraction (Asbridge *et al.*, 2013).

Mobile phones also emit radio frequency electromagnetic radiation. It is non-ionizing radiation but they are likely absorbed by the tissues which may cause any tissue heating effect which may cause ear discomfort etc. Mobile phones transmit radiations to transforms through radio frequencies (Myoung Soo Kwon *et al.*, 2011). Radio frequencies are not harm ionising radiations like x-rays and gamma rays. But when these radiations from the antenna of the mobile phones may cause effects on continuous transmission. It may lead to tumour in that particular area. It is likely to have children more than adults as they have thinner skull bone it is easy to transmit these rays towards the body. It is not evident that these radiations cause cancer but on over usage, it may lead to the formation of tumour (Rubin *et al.*, 2005).

In earlier days people use telephone to communicate with people. At that time, it has so many restrictions related to timing and charge for the phone call so people will talk only if it is necessary and important to communicate message. Most of the people prefer letter to pass the information to others. Now a days there is no such restriction for time and cost. Cellular network is enough to communicate with people through call or video call, which will make people to spend more time on chatting and video call.

Many studies have reported that there is a negative effect of sleeping pattern in the extensive usage of mobile phones. Smartphones are portable, hand-held devices that have much higher potential of interrupting sleep quality or quantity (Carter *et al.*, 2016). Problematic smartphone use has been consistently linked to poor sleep-in previous studies, and smartphone overuse has been associated with daytime tiredness, longer sleep latency, and reduced sleep duration (Chung *et al.*, 2018), (Chindamo *et al.*, 2019) In particular, smartphone use close to

sleep initiation has been shown to delay circadian rhythm and found associated with total sleep time, where longer usage was associated with poor sleep. However, despite consistent advice from health bodies concerning the negative impacts of smartphone use on sleep, the majority of adults use their phones during the night and close to bed time (Alshobaili *et al.*, 2019). When mobile phones are kept close to our head unless in airplane mode some apps will be working on it where it radiates some frequencies lesser than the normal usage. And also, when mobile phones are kept near our head will stimulate the urge to use it continuously, will make us to keep on checking it frequently, will make us to scroll social network. This will highly disturb our sleep and make us to feel tired in the daytime.

Children have the potential to be at greater risk than adults for developing brain cancer from cell phones. Their nervous systems are still developing and therefore more vulnerable to factors that may cause cancer. (Ryan Gorman., 2011).

Smart phones (i.e., phones with the ability to download and run applications) become necessities of younger adult (Smith, 2012). A recent international systematic review found that the prevalence of smartphone addiction was around 25% in teenagers and young people (Sohn *et al.*,2019). Through smart phones, younger adults often download a lot of applications; however, they quickly abandon or rarely use most of them. Smart phones offer many useful functions to improve older adults' quality of life; however, such new functions are rarely being used by older adults. This is the problem that practitioners face. They design elderly specific tools and services (e.g., global positioning system tracking and the medication reminder) with utility, but the utility is in vain if older adults refuse to try new functions (Zhou *et al.*,2014)

New functions could be intimidating for older adults because older adults used to experience difficulties in using mobile phones (e.g., hierarchical menus, poor readability, and compressed keys). To avoid these problems, older adults used limited functions (mainly phone calls and short messages). As a result, many older adults still stick to feature phones (i.e., phones without the ability to download and run applications). (Zhang, 2009)

Connectivity is one of the influencing factors for the usage of mobile phone (Samoek, 2018). It is available in the form of WIFI, mobile data, Bluetooth, USB, MODEM etc. If Wi-Fi is available people tend to use mobile phone for a longer period. Recent studies have also reported the increase of mobile phone dependence, and this could increase internet addiction (Billieux *et al.*, 2008). Surfing data in a smaller screen is difficult for elder people as the font

size and screen is smaller, understanding, recognition. Younger people prefer to have mobile phone in small font and less brightness (Jia Zhou, 2014).

Biomarkers of oxidative stress can be classified as molecules that are modified by interactions with ROS in the microenvironment; and molecules of the antioxidant system that change in response to increased redox stress. DNA, lipids (including phospholipids), proteins and carbohydrates are examples of molecules that can be modified by excessive ROS in vivo. The most promising biomarkers are the ones that closely correlate with the pathophysiological process of the disease (Griendling *et al.*, 2003), (Harrison *et al* 2005), The role of oxidative stress in the pathophysiology of CVD is well established. Reactive oxygen species (ROS) are derived from many sources including mitochondria, xanthine oxidase, uncoupled nitric oxide synthases and NADPH oxidase (Mueller *et al.*, 2005)

Oxidative stress is defined as the imbalance in the redox characteristics of some cellular environment which can be the result of either biochemical processes leading to the production of reactive species, exposure to damaging agents (i.e., environmental pollutants and radiations), or limited capabilities of endogenous antioxidant systems. Reactive oxygen and nitrogen species (ROS/RNS) produced under oxidative stress are known to damage all cellular biomolecules (lipids, sugars, proteins, and polynucleotides). To counteract reactive oxygen species–induced cell damage, biological systems have evolved endogenous mechanisms to protect themselves in normal physiologic conditions (Valko *et al.*, 2007).

The cellular antioxidant mechanisms involve nonenzymatic compounds and enzymes such as superoxide dismutase (SOD), catalase, and glutathione peroxidase (GPx). (Trachottam *et al.*, 2007) Superoxide dismutase is specific to superoxide-radical detoxification. Superoxide dismutase can rapidly dismutase the superoxide radical, yielding hydrogen peroxide and oxygen. (Vaculin *et al.*,2010) Hydrogen peroxide is a diffusible reactive oxygen species that contributes to the development of pathologic pain states, not only by generating harmful reactive species but also by modulating synaptic plasticity. (Takahashi *et al.*,2007). The presence of hydrogen peroxide apparently affects the release of intracellular calcium, leading to neuronal sensitization and pronociceptive patterns in interneurons in the spinal cord dorsal horn. Catalase activity, in turn, converts hydrogen peroxide to water. (Vaschulin *et al.*,2010), (Negre-Salvayre. *et al* 2010).

During the past decade, research has revealed a widespread involvement of oxidative stress in a number of disease processes, including cancer, cardiovascular disease (CVD),

atherosclerosis, diabetes, arthritis, neurodegenerative disorders, and pulmonary, renal, and hepatic diseases (Valko *et al.*, 2010), since then usage has increased. Smartphone ownership in children aged 11 and older is ubiquitous, and the prevalence of mental health problems peaks during the teenager years. The majority of the young adults reported that they were expected to be reachable via the mobile phone all day or around the clock (Sara Thomee *et al.*, 2011.)

Smartphone addiction is prevalent and occurs more frequently amongst younger adults. Proxy measures of screen time were not synonymous with addiction; a validated addiction instrument should be used to capture this phenomenon. Those exhibiting smartphone addiction experienced poorer sleep. Mobile phone is becoming an integral part to students with regard to managing critical situations and maintaining social relationships (Roberts *et al.*, 2014).

Therefore, this study aims to understand the above-mentioned difference in mobile phone use and this study examined age-related differences in requirements for mobile phones in the context of the transition from feature phones to smart phones.

the following objectives were formulated to meet the goal of the study

Objectives of the study:

- To explore the impact of extensive usage of smartphone users through online questionnaire.
- To evaluate oxidative biomarkers in the blood sample of over-usage of smartphone users.
- To determine the levels of oxidative marker related to age among extensive usage of smartphone users.

The study was conducted in two phases. In phase I, exploration of impact of extensive usage of mobile phone through online questionnaire was carried out to know whether on extensive usage of mobile phones cause any effects to human, where questionnaire was framed carrying the demographic details, habituation, awareness questions and questions related to occurrence of pain. In phase II Blood samples were collected from study volunteers and level of oxidative biomarkers were estimated. Statistical analysis was carried out using SPSS 25.0 software to find the correlation of oxidative stress on extensive usage of mobile phones.

Review of literature

REVIEW OF LITERATURE

The literature pertaining to the present study on “**Evaluation of oxidative biomarkers in relation with the self-reported symptoms among extensive usage of smartphone users on basis of Age**” is reviewed under the following headings:

2.1 INTRODUCTION

2.2 SMART PHONE USAGE

2.3 POSTURE OF MOBILE USAGE

2.4 EFFECT ON SLEEP

2.5 ADDICTION BEHAVIOUR AND MENTAL ILLNESSES

2.6 MOBILEPHONE RADIATION

2.7 PSYCHOLOGICAL STRESS

2.8 EAR PROBLEM

2.9 MYOPIA

2.10 CANCER RISK

2.11 ORGAN RISK

2.12 MUSCULOSKELETAL COMPLAINS/ DISORDERS

2.13 CORRELATION BETWEEN MUSCULOSKELETAL DISORDER AND LEVELS OF ERGONOMIC RISK AMONG SMARTPHONE USERS

2.14 MOBILE PHONE USAGE IN RELATION WITH AGE

2.1 INTRODUCTION

Mobile phones are powerful communication devices, first demonstrated by Motorola in 1973, and made commercially available from 1984. Now those Portable devices became integral part of our lives. Mobile phone is become as the most dominant portal for information and communication technology. A mental impairment resulting has come to the attention of sociologists, psychologists, and scholars of education on mobile addiction. Over time usage of Mobile phone may increase anger, tension, depression, irritability, and restlessness which may

alter the physiological behaviour and reduce work efficacy (Subramani Parasuraman *et.al.*,2017).

Now a days increase in use of smartphones among children and young people (CYP) which has occurred at the same time as a rise in common mental disorders in the same age group, including reported depressive symptoms, poor sleep and suicide ideation with grave implications for life-long mental health and the healthcare economy. There is well fare in UK for the CYP which tells us between the association of mobile phones usage among the children. The study explains the challenge in which the study is on harmfulness due to screen time of mobiles but the result was not accurate due to other reasons such that the person may gone through other electronic devise such as television. Recent years have seen an explosion in research considering the prevalence of problematic smartphone use (PSU), which has been operationalised in such a way that it maps onto concepts of behavioural addiction: tolerance, withdrawal, preoccupation, neglect of other activities, subjective loss of control and continued use despite evidence of harm. Other behavioural addictions, such as problem gambling, show robust associations with common mental disorders such as depression, where sporadic gambling does not. If a distinctive problematic pattern of smart phone use can be demonstrated to be prevalent, and if this pattern of use is associated with harm, there is value in identifying children and young people with this pattern of use and potentially addressing it clinically (Sei You Sohn.*et.al.*,2019).

Smartphone has used more widely over the past 1 decade. There are emerging reports of problematic behaviour patterns in relation to smartphone. Smartphone addiction describe a decreased interest in face-to-face relationship. Measuring duration of use is an inexact proxy for addiction, as some people may experience the features of addiction with a lower duration of use while others may use their phone in an adaptive way for long periods of time (for example, answering work emails during a long commute) but be able to put the phone down without distress and attend to appropriate activities such as communicating with family members or going to bed on time. While the negative effects of screen time make higher potential of interrupting sleep quality or quantity. Smartphone overuse has been associated with daytime tiredness, and reduced sleep duration. In particular, smartphone use close to sleep initiation has been shown to delay circadian rhythm and found associated with total sleep time, where longer- usage was associated with poor sleep. Furthermore, poor sleep outcomes may

mediate the relationships between smartphone addiction and behaviour (Stale Pallesen *et.al*, 2020).

The term ‘addiction’ is defined with a certain activity that results in a disturbance of his/her daily activities and shows a pattern similar to substance dependence. The recent studies also reported the increase of mobile phone dependence, and this could increase internet addiction. Over usage of mobile phones may cause psychological illnesses such as dry eyes, computer vision syndrome, weakness of thumb and wrist, neck pain (Subramani Parasuraman *et al.*, 2017). Frequency of mobile phone use, the purpose of mobile use, the degree of neck flexion while using phone, and the body position are the factors associated with neck and shoulder pain and its severity.

2.2 SMART PHONE USAGE

The mobile phone technology has brought the world closer. It provided convenience in communication among people by way of calling or texting. The mobile phones are coming up with variety of features like internet access, sending e-mails, games, access to social networking sites like face-book, listening to music, playing radio, reading books, dictionary and so on. The mobile phones are also used to overcome the feeling of loneliness. The majority of the users are in the age group of 15 to 25 years. The contacts are established instantly with the help of mobile phones which was not possible earlier. However, though the mobile phone provided many advantages, it has also caused some problems also. Some people are using the mobile phones so excessively that it assumes the form of addiction. The use of mobile phones has reduced the face-to-face communication. It is observed that the people sending text messages while talking to others. The students are using mobile phones for playing games, sending messages, calling even when the class is in progress. The mobile phones are used at places like hospitals, judicial courts, petrol pumps where their use is banned. The mobile phone use during driving is commonly observed which may increase the chances of involving in accidents. The use of mobile phone while driving can distract attention of the driver visually, physically and cognitively. Earlier studies have shown that various personality traits like neuroticism, extraversion, psychoticism, etc. and mobile use have some relation to each other. The gender may also play some role in high mobile phone use (Alpana Vaidya. *et al.*,2016).

Mobile phone addiction and withdrawal from mobile network may increase anger, tension, depression, irritability, and restlessness which may alter the physiological behaviour and reduce work efficacy. (Subramani Parasuraman *et.al.*,2017).

Problematic smartphone usage (PSU) has been consistently associated with measures of poor mental health, in particular relating to depression, anxiety, stress, poor sleep quality. There are some signs of smartphone addiction are constantly checking the phone for no reason, feeling anxious or restless without the phone, waking up in the middle of night to check the mobile and communication updates, delay in professional/ Institution performance as a result of prolonged phone activities, and distracted with smartphone applications (Subramani Parasuraman, *et.al.*,2017).

Excessive use of mobile phones is paired with the negative effect on human. It creates dependence on the gadget may increase the level of anxiety and depression Rosen et al., 2013. Jones., 2014 conducted an online survey and found that students are addicted more towards mobile phone which may cause serious negative psychological effects. Boumosleh & (Jaalouk., 2017) investigated whether anxiety and depression independently led to smartphone addiction. Their sample was 668 random Lebanese undergraduate students. 668 Lebanese undergraduate students were chosen at random. Depression and anxiety were also found to be positive predictors of smartphone addiction in their cross-sectional investigation. They also discovered that depression levels were a stronger predictor than anxiety ratings.

2.3 POSTURE OF MOBILE USAGE

The smartphone users tend to report pain in the neck, shoulder, and thumb, and the severity of the symptoms increases as the total time spent using the smartphone (Sang in Jung, 2016). Prolonged smart phone usage causes faulty posture such as forward neck posture or rounded shoulders. Sustained forward neck posture can cause injury to the structure of the cervical and lumbar spine, as well as ligaments. These structural problems caused by posture can also lead to respiratory dysfunction. People tend to have comfortable tilted head neck position for a long period of time unknowingly which in turn cause serious effects. This is because of too much addiction towards the mobile phone. In other case, people who work in software company or any other related field have to sit in front of laptop for a about 8 hours for work. This will also cause serious effect in the neck and spine position. Kim, 2015 studied about the association of smartphone usage and neck pain, where he reported that smartphone

users had neck pain due to the increased cervical angle. Gak *et al.*, 2013 reported that prolonged sitting and using of smartphone cause rounded shoulder which is associated with FHP (Forward Head Position). FHP is caused when a person leaning their head forward which is out of neutral alignment with their spine.

Perry *et al.*,2004 investigated the possibility correlation between dysfunctional breathing and musculoskeletal pain patterns. Their result showed that 83% of patients with neck pain (caused by faulty posture) experienced a changed breathing pattern. This study indicated that there was a relationship between neck pain and respiration. The adverse effects of prolonged sitting and a sedentary lifestyle on our health are well known, and similarly, we should understand the effect of prolonged smartphone usage on our posture and respiratory function. (Perry *et al.*,2004)

2.4 EFFECT ON SLEEP

Sleep plays an important role in our body. It regulates the physical functions, cellular toxin removal, disease prevention, and restoration of both the mind and body (Curcio *et al.*, 2015). Proper sleep is important for all age people. Owens (2014) states that lack of sleep-in adolescents is becoming an important health issue worldwide. Current metanalysis by Carter et al (2016) showed that bedtime use of media devices was positively associated with poor sleep quality and excessive daytime sleepiness. In most of the house lights will be switched off after 10pm. Mobile phones while used at bed time with no lights may leads to poor quality sleep by various mechanisms (Mireku *et al.*, 2019). Because of the revolution in the technology people can access anything at the same place. This enables them to use the internet, social media, video calls, chatting, playing games etc for a longer period of time. Major factor which affects human is the blue light from the screen of mobile phones. Chang *et al.*, (2015) states that this blue light from mobile screen can cause decrease in the production of melatonin. Melatonin is a hormone which controls the sleep/wake cycle or circadian rhythm. So, reduction in melatonin cause difficulties to fall and stay asleep.

Nazish Rafique *et al.*, 2020 reported that using mobile phones after lights off for at least 30 minutes can leads to poor sleep quality, daytime sleepiness, sleep disturbances and increased sleep latency. And also, sometimes placing mobile phones near the head will stimulate some urge to check mobile phone frequently, reply to messages will also have effect on sleep.

2.5 ADDICTION BEHAVIOUR AND MENTAL ILLNESS

Rapid development in the technology as created so many gadgets where smartphones play a major role. From younger people to old people are now of mobile phones. According to the quote too much of anything is good for nothing, people now have started to use mobile phone most of the time. It has now become a part of a body. Mobile addiction makes people too dependent on it. Notably younger people now tend to use mobile phones for the whole day. They keep on checking it which itself an addiction behaviour. Youngsters now using mobile phones widely at the time of sleep which will leads to mental disturbances, anxiety, depression. Schoeni et al., (2015) reports that mobile phone usage during night hours was common among youngsters and reported that poor perceived health was shown due to staying up all night. No recordable association was found between memory performance and mobile phones. Boumosleh & Jaalouk (2017) investigated whether anxiety and depression independently contributed to smartphone addiction. Their sample was 668 random Lebanese undergraduate students. Their cross-sectional study proposed that depression and anxiety were also a positive predictor of smartphone addiction. They also revealed that with depression scores were a more powerful predictor as compared to anxiety.

There are issues specific to the population with serious mental illness that could impede smartphone use. People with serious mental illness have conditions that are often accompanied by socioeconomic disadvantage, limited education, symptoms, and cognitive impairments that could affect phone use. The rapid increase in the rate of smartphone use has also occurred in the population with serious mental illness, with growth trends and ownership rates comparable to the general population. here have been studies of mobile phone ownership in people with serious mental illness (Firth et al., 2015)

2.6 MOBILE PHONE RADIATION

Mobile phones transmit signals to nearby cell towers using Radio Frequency waves. This phones work on several frequencies in Radio Frequency bands from 850 MHz to 1900 MHz which correlate with wavelengths in air 35 and 15 cm. These frequencies are close to the frequencies with are employed in the microwave and some other medical equipment used in hospitals. This is a kind of energy in the electromagnetic spectrum like FM radio waves. Like radio waves, visible light, heat, microwaves, Radio Frequencies waves are a kind of non-ionizing radiation. These waves are not capable of causing cancer directly by damaging the

DNA. These Radio Frequencies are different from stronger radiations like X-rays, gamma rays, UV rays which is capable of causing mutation leads to cancer.

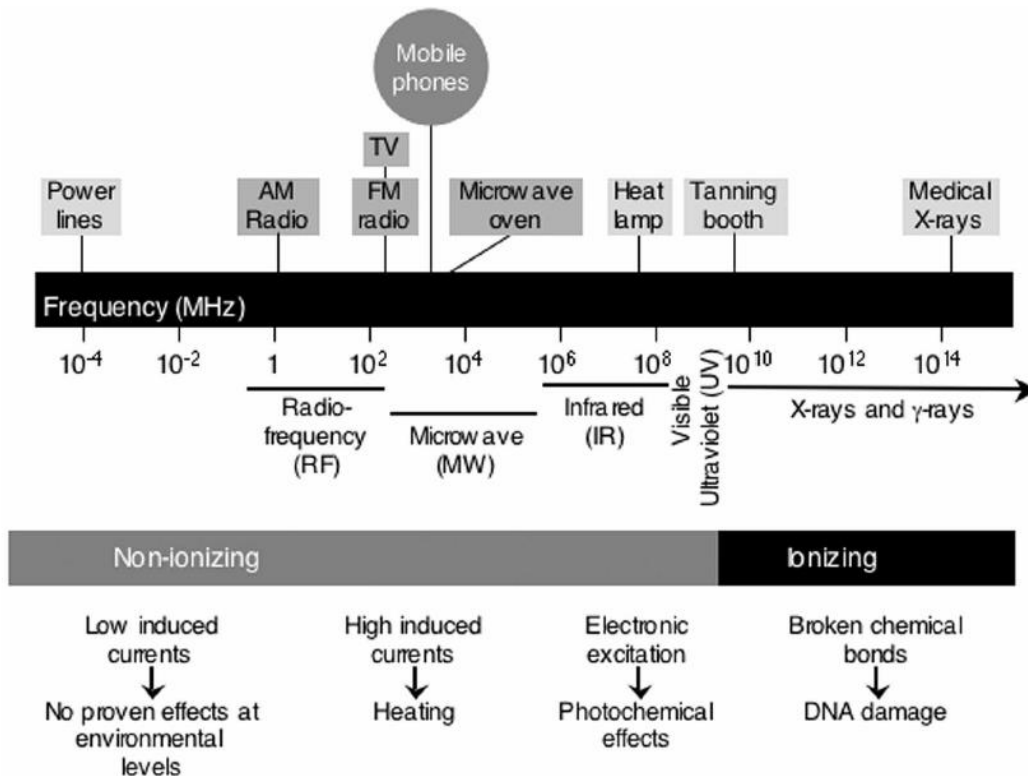


Figure 1 ELECTROMAGNETIC SPECTRUM

Foster & Moulder (2000).

Radio Frequency absorbed by the body of mobile phone user can be calculated by the Specific Absorption Rate (SAR). It is calculated from mobile phones antenna which produce RF waves. The waves are most likely to affect the head as it is held closer while talking to people. The body tissue closer to the phone absorbs more energy from Radio Frequency waves than the tissue which is far away. Many factors affect the person when they are exposed to Radio Frequency waves like using mobile phone for a longer period of time, keeping mobile phones near to head while sleeping, when people are using mobile phones closer to head instead of having speaker mode, amount of cell phone traffic in a particular time, model of mobile phone being used as each phone has made in different model, path and distance to the nearest cell phone towers. Putting the mobile near pillow while sleeping caused increased sleep latency, sleep disturbances and daytime sleepiness. These effects may be caused by a continuous urge to see notifications and updates on the nearby placed phone, disturbance created by the vibrations from receiving notifications and messages, heat generated by charging phones and RF-EMF exposure from the mobile phone (Sahin *et al.*, 2013). Because mobile

phones are held near to the head while talking to people there may be some risk of causing tumour in that area. But there is no proper research that these frequencies cause cancer.

They may cause cancer to human on over exposure of radiation and heating effect. During the sleep time, when the mobile phones are not in use, they still emit RFEMFs; however, the levels are much lower than that of a phone call. Moreover, the smart phones are constantly scanning for signals, text updates, emails, and software updates, app updates. Even notifications that we receive through apps require a certain level of radiation to be released (Bosquillon *et al.*, 2020). When mobile data/ WIFI switched on while sleeping several applications will be working in the background. It is reported the media coverage of a case in which a Florida man claimed that his wife died of brain cancer caused because of the hand-held mobile phone position (Foster & Moulder 2000).

2.7 PSYCHOLOGICAL STRESS

Neuroscience Institute in Manhasset, New York, considered that there is no solid proof between cell phone radiation and tumour risk but the possibility still may exist. Adolescents are at high risk of being smartphone addicts (Cha and Seo, 2018). Excessive use of smartphone makes negative attitude and feeling of anxiety and dependency on may increase the risk of anxiety and depression (Rosen et al., 2013, Thomée *et al.*, 2011).

Jones (2014) conducted an online survey and found that students seemed to be addicted to their mobile phones. It was concluded that the excessive smartphone use had a negative psychological effect. On prolonged usage of mobile phone, it leads to some external problem like sleeplessness, insomnia, depression, anxiety etc. A range of different personality and emotional factors were investigated in relation to PSU (Problematic smart phone usage). Some traits were associated with greater risk-taking (such as low self-control, impulsivity, emotional instability) and traits associated with avoidance of risk taking, were more common amongst problematic smartphone users (Sei Yon Sohn *et al.*, 2019)

2.8 EAR PROBLEM

Earlier days telephone was used to communicate with people, where it has limitations so people use it for important purpose alone to convey the message. As technology develops, in a small gadget called smart phone with internet facilities we can now talk with people for a longer period time without any time limitations and cost limitations. Velayutham *et al.*, (2011)

reports that there was unilateral hearing loss in the dominant ear implies that the mobile phone as the cause of hearing loss. There appears to be correlation between the duration of usage of mobile daily and the length of years using mobile phone to the degree of hearing loss. The operating frequencies of most mobile phone are around 900–1800 MHz. Although this frequency is within the acceptable range yet data on its long-term cumulative effect and influence on cellular damage is lacking. The ear, remain closest to the mobile phone and is the direct recipient of the noise, thermal energy, and EMR waves emitted by the phone (Uloziene *et al.*, 2005). But on prolonged talking through mobile phone can cause adverse condition by causing heating effect.

2.9 MYOPIA

Smartphones, iPads, tablets and computers are used at a very early age in both home and school environments (Ayaki *et al.*, 2016). Children and elder people are very much dependent on mobile phones as it has all the features like songs, books, social media etc. Excessive use of digital smart devices, including smartphones and tablet etc could be a risk factor for myopia. The widespread of myopia is increasing worldwide, with half of the global population expected to have myopia by 2050. Along with myopia and it also causes irreversible blindness may occur in upcoming decades. The myopia epidemic is likely to be driven by exposure to environmental risk factors present in ever more urban and developed societies, with two major risk factors of particular concern: insufficient time spent outdoors and more time engaged in so-called near-vision. (Joshua Foreman, *et.al.*,2021)

McCarran, Loughman, Butler *et al.* 2020 have conducted a survey among students from primary school, secondary school, and University. It is reported that students are using mobile phones at an average rate of four hours a day. 73% of people believed that digital technology adversely affects the eyes. Students also reported that over screen time cause eye strain, dry eyes, headaches, difficulty in reading, and 31 percent reported myopia. The escalating prevalence of myopia is not a recent phenomenon and certainly predates smartphones, but the current generation of children are the first to grow up in an era of smartphone dependency. During pandemic the whole world is dependent on mobile phone for to know information, for education, to communicate with people, to play games as no one is allowed to go out and be in a gathering. At the same time the risk was also high on over usage of mobile phones.

2.10 CANCER RISK

Ionising radiations are the key factors for causing brain tumours. Glioma is the most common malignant tumour which represents about 60% of all nervous system tumours. Certain studies have been conducted to see the relation between mobile phone usage and cancer. In Amritsar, Punjab, India, a study was conducted to see the relation between mobile phone use, and DNA and chromosomal damage in lymphocytes of mobile phones users were observed. Such damages can have long-term consequences in terms of increased risk of neoplasia or other age-related changes (Volkow *et al.*, 2007). Children has the smaller head and thinner skull bone than adults, so it is easy to absorb the radio frequencies easily. But there is no scientific proof by the researchers that mobile phones can cause cancer. At the same time excessive use of mobile phones should be avoided and cautious to prevent cancer.

2.11 ORGAN RISK

Nowadays social media has created a serious impact on humans but sticking to it all the time. This addiction leads to prolonged use of mobile phones which may cause serious psychological effects like eyes dryness, eyesight problems, wrist and thumb pain, muscular pain, neck pain, insomnia, delusions, sleep disturbance, stress, hallucination, lower self-confidence, increased frequency of De Quervain's tenosynovitis, etc. (Parasuraman *et al* 2017).

Mobile phones provide heat on over-usage which may cause adverse health effect which is reported in a study that when mobile phones are kept close to the chest it increases the heart rate and decreases when kept close to the head, but this is not convincing in comparison to conditions without using mobile phone. But again, researchers haven't suggested any valid evidence of adverse health effects from exposure to radiofrequency fields except the tissue heating effect (Roosli, 2008).

2.12 MUSCULOSKELETAL COMPLAINS/ DISORDERS

Over-usage of mobile phones will be of different angles and types where head angle will be tilted, change in the wrist, thumb and hand movement, neck position which will create pain. This depends on the task, posture, and position of holding the smartphone. Several studies have suggested that neck pain caused while using a mobile phone will make us adopt a flexed position which causes a negative effect on musculoskeletal problems. There is a huge difference in using a mobile phone while sitting and standing. The head-neck angle shift is higher in a sitting position than that while standing. (Eitivipart *et al.*, 2018). The faulty posture

of the head and neck will cause serious effects in the ligament alignment, cervical pain, and respiration dysfunction (Jung *et al.*,2018)

2.13 CORRELATION BETWEEN MUSCULOSKELETAL DISORDER AND LEVELS OF ERGONOMIC RISK AMONG SMARTPHONE USERS

While using mobile phones there will some flexed position like neck, shoulder, elbow, wrist, hand flexion when typing, with their hand and wrist to support the phone while upper and lower back, hip and thigh and ankle and foot neutral was also flexed (Namwongsa *et al.*-2017). This position adopted while using mobile phones will surely make adverse effect in the body (Lee *et al.*, 2014). People tend to be in same position for a long time or they adapt different position for the sophisticated usage of mobile phones. If the awkward position continues it will cause strain in the muscle and make change in the ligaments around the joint also nerve issues. Such positions like head-neck tilted angle, arm raising can lead to ergonomic problems which affect the ergonomics risk (Chaiklieng *et al.*, 2016). From this we can understand there is a significant correlation between the smartphone usage and musculoskeletal disorder may cause due to flexed awkward position which leads to the increase in the levels of ergonomic risk.

2.15 MOBILE PHONE USAGE IN RELATION WITH AGE

Mobile phone development has led to a condition where it is considered ubiquitous among the population. Even older people are now upgrading themselves to adopt to this technology. Older people are now likely to have a mobile phone than desktop or computer. Most of the people are now likely to note data and information in mobile phones than writing in a notepad or diary. Most of the older people after retirement they need something to pass their time. While using mobile phone most of the time if occupied for them. New functions, design and working pattern of mobile phones is quite difficult for older people than younger people. Younger people adapt the steps to access mobile phone easily while elder people will strain to remember the steps involved in to access the mobile phones. But somehow older people are trying to get adapted to these steps. Age-related differences in vision are well documented. Older adults preferred larger display, larger font, and larger icon to read information clearly. They experienced greater difficulties than younger adults in the perception of mobile devices such as mobile phones, personal digital assistants (PDA), and digital diaries. (Lee, *et al* 2007). Older people use mobile phone in larger font and in high brightness mode.

Where most of the younger people will have smaller font and in less brightness mode. Where older people are more radiated to the blue light of the mobile phone which may leads to eye problems easily (Zhou *et al.*, 2012), (Renaud, *et al.*, 2008), (Van Biljon *et al.*, 2008).

Connectivity was one influential factor of younger adults' perception of mobile phone usability. When choosing mobile phones, they would consider the connectivity options available through GPRS/USB/Infrared/ Bluetooth/Wi-Fi (Zhang, 2009). Browsing in small screen is difficult for older people as the screen would be small and they feel some problems like visibility, focus recognition, understanding, operation of web page structure, access button, underlined hyperlinks etc. Younger people prefer this handy thing and use online shopping, download PDFs and work on it as they feel this as comfortable. Both the younger and older people perceived privacy equally important (Jia Zhou *et al.*, 2014). Difficulties with smart phones that younger adults encounter now may be encountered by older adults in the future when they use smart phones. For example, older adults use limited functions, so finding the desired function among limited functions can be easy. However, as their experience grows and they use more functions, finding the desired function among a large number of functions could be more difficult, so they would stress the importance of ease to find the desired function, which is similar to younger adults. Younger people are easy to understand the manuals, font size, icon size and operations than old people.

Experimental Procedure

EXPERIMENTAL PROCEDURE

Smartphone use has become ubiquitous over the past decade. This has been accompanied by growing concerns around excessive and potentially harmful use. There are emerging reports of problematic behaviour patterns in relation to smartphone use which mirror those of addiction. Over usage of mobile phones may cause psychological illness such as dry eyes, computer vision syndrome, weakness of thumb and wrist, neck pain and rigidity, increased frequency of De Quervain's tenosynovitis, tactile hallucinations, nomophobia, insecurity, delusions, auditory sleep disturbances, insomnia, hallucinations, lower self-confidence, and mobile phone addiction disorders (Peraman *et al*, 2016). Hence this study aimed to assess the level of oxidative biomarkers among users on extensive usage of mobile phone and to explore the with self-reported symptoms of participants and its associated complications.

As a preliminary study, an online survey was conducted between November 2021-December 2021. Questionnaire was prepared and online survey method was carried out to collect the data including demographic details, habituation, previous awareness on mobile phone hazards and addiction behaviour of the participants. Number of participants attended for the study is 100.

Statistical analysis was performed using SPSS 25 to analyse the data of participants based on the age and the results were summarized.

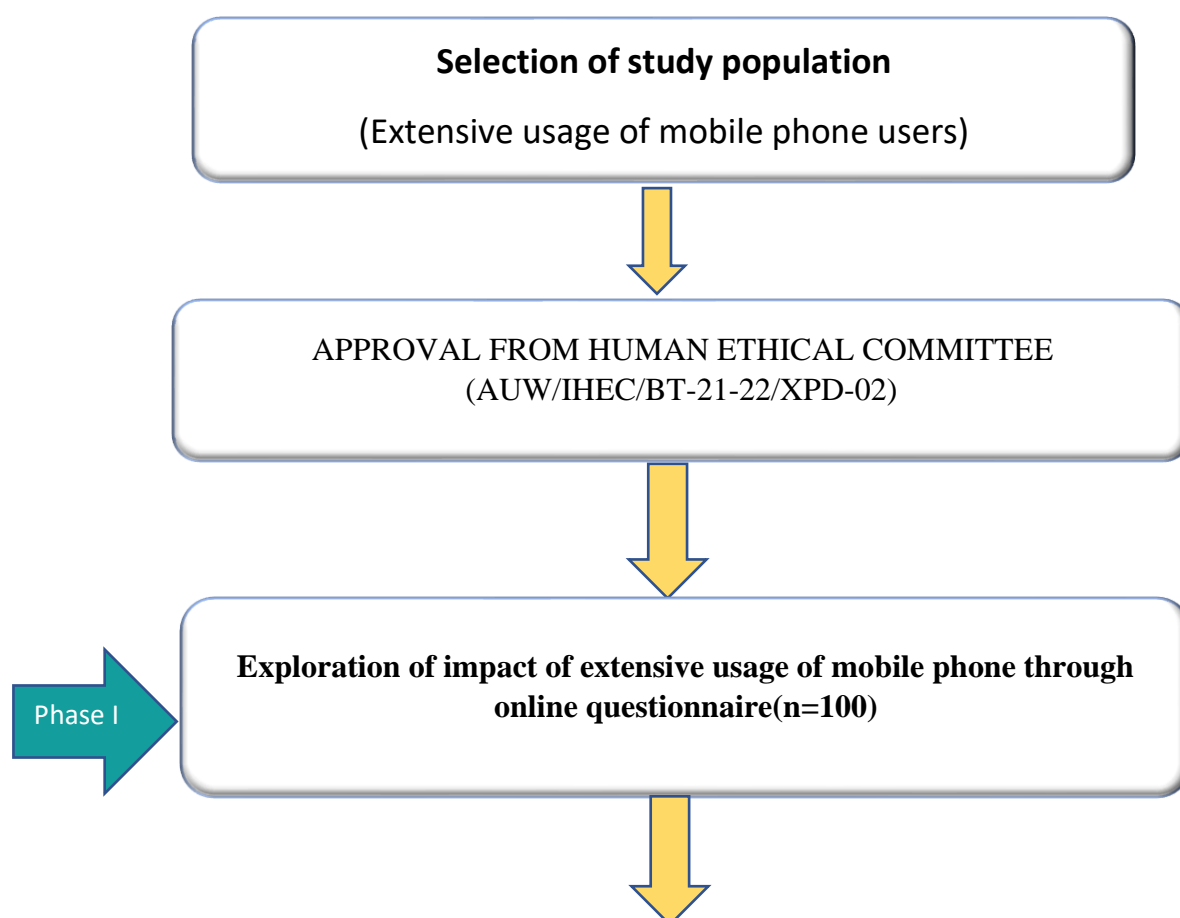
Oxidative stress reflects an imbalance between the systemic manifestation of reactive oxygen species and the level of antioxidants defense system. In humans, Oxidative stress is thought to be involved in the development of cancer, Parkinson's disease, atherosclerosis, heart failure and myocardial infarction (Devasagayam, 2004). The Oxidative damage is tested by the increase in lipid peroxidation formation.

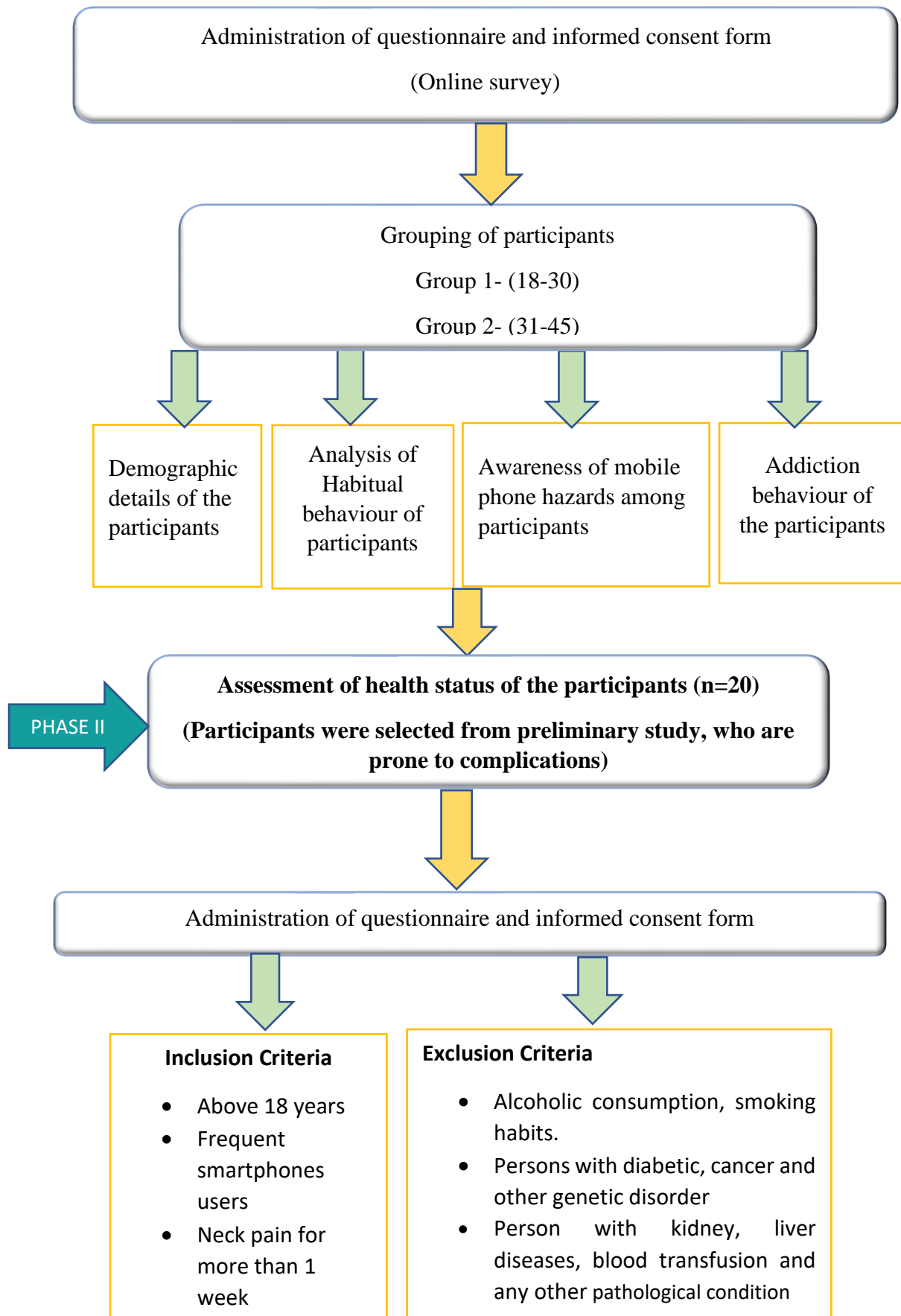
Among the cellular molecules, Lipids contain more than one double bond susceptible to action of free radicals. The resulting reaction is known as lipid peroxidation which produces a large number of by products being studied in relation to disease. These by products being studied in relation to diseases. These by products can be measured by the estimation of aldehyde products using thiobarbituric acid (Devasagayam, 2003). Heavy metals induce an increased production of malondialdehyde (MDA), an indicator of lipid peroxidation and whole body free radical activity (Flora *et al.*, 2008). High levels of lipid peroxides are associated with cancer, heart disease, stroke and adding.

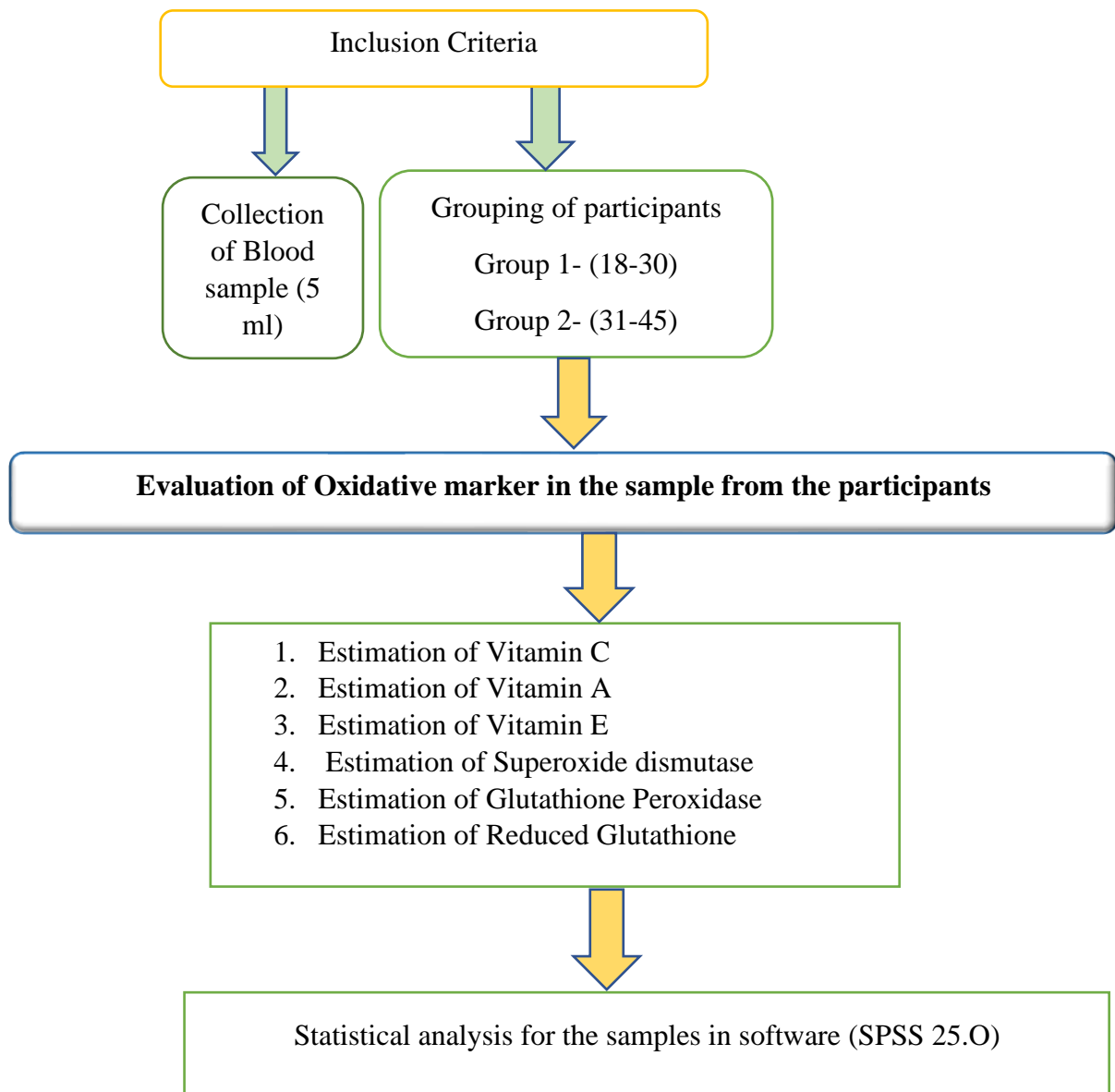
Antioxidants are the compounds that protect the body against toxic effect of free radicals. The protective mechanisms of antioxidants serve to scavenge the free radicals. Different antioxidants act at different levels. They may prevent initiation of chain reaction by removing free radicals (preventive Antioxidants). They may scavenge free radicals generated in chain reaction, there by interrupting the chain sequences (chain breaking). They may remove peroxides, thereby preventing further generation of reactive oxygen species (ROS). Types of antioxidant system such as Enzymes antioxidant system, Vitamins antioxidant system, Minerals antioxidant system. (Sei You Sohn *et al.*,2019).

Based on the above facts, Evaluation of oxidative biomarkers was conducted in peripheral blood plasma to find out the relationship between the self-reported symptoms on extensive use of mobile phones with the level of oxidative biomarkers.

WORK PLAN:







The Experimental design adopted in the study “Evaluation of oxidative biomarkers in relation with the self-reported symptoms among extensive usage of smart phone users on the basis of Age” was conducted in two phases and it is discussed under the following headings:

PHASE I

3.1 EXPLORATION OF IMPACT OF EXTENSIVE USAGE OF MOBILE PHONE THROUGH ONLINE QUESTIONNAIRE

3.1.1 Survey conducted through online among participants based on Age

3.1.2 Demographic profile of the participants

3.1.3 Analysis of Habitual Behaviour of participants

3.1.4 Addiction Behaviour of the participants

3.1.5 Awareness of mobile phone hazards among participants

PHASE II

3.2 EVALUATION OF OXIDATIVE MARKERS OF MOBILE PHONE USERS

3.2.1 Study population

3.2.2 Administration of questionnaire and

3.2.3 Collection of blood sample

informed consent form

3.2.4 Grouping of participants based on Age

3.2.5 Evaluation of Oxidative markers in the sample from participants

3.2.5.1 Estimation of superoxide dismutase

3.2.5.2 Estimation of Glutathione peroxidase

3.2.5.3 Estimation of Reduced glutathione

3.2.5.4 Estimation of Vitamin A

3.2.5.5 Estimation of Vitamin E

3.2.5.6 Estimation of Vitamin C

3.2.6 Statistical analysis

PHASE I

3.1 EXPLORATION OF IMPACT OF EXTENSIVE USAGE OF MOBILE PHONE THROUGH ONLINE QUESTIONNAIRE

Questionnaire was prepared in the form of google form which was composed of 36 questions self-designed by the investigators. The link of the questionnaire was forward to people through social media to the contacts of investigator. Participants were encouraged to roll of questionnaire to as many contacts as taken.

3.1.1 Survey conducted through online among participants based on Age

Questionnaire is prepared and shared among the participants. Participants are divided into two groups as Group 1 (18-30) and Group 2 (31-45). Conducted in the form of online survey among the participants. (Annexure 1)

The following information were obtained from the participants:

3.1.2 Demographic profile of the participants

Demographic details talk about the detailed note on age, gender, academic qualification, accommodation, Occupation of the participants. It collects the personal details of the people.

3.1.3 Analysis of Habitual behaviour of participants

Habituation details talks about the years of mobile phones usage, duration of mobile phone usage per day, posture of mobile phone usage and also involve the medications details

3.1.4 Addiction Behaviour of the participants

Addiction behaviour collects details about how participants are addicted towards mobile phone. It is a kind of self-analysis of how participants are addicted. It includes questions like how much time they spend on mobile phone, frequency of checking mobile phones, missing social life.

3.1.5 Awareness of mobile phone hazards among participants

Analysis of awareness gives detailed information about how people are aware of mobile phone usage ill effects. It includes questions like awareness on EMR, neck pain, ear discomfort.



3.2 EVALUATION OF THE HEALTH STATUS AND OXIDATIVE MARKERS OF MOBILE PHONE USERS

Google form was created to collect data from the participants in which physical and mental health status of participants were determined (n=100). On extensive usage of mobile phone, it is observed that it leads to many health complications.

From the preliminary study specific participants those who are prone to complications on over usage of mobile phones are selected for the study (n=20). Those participants sample are collected oxidative biomarkers are determined.

3.2.1 STUDY POPULATION

A cross sectional study was conducted among the participants in (Renga nagar, Periyankenplayam Coimbatore), Samples along with details of participants are taken from the Renga nagar area with the permission of area counsellor Mr.J. Pon Madasamy, on basis of the Age.

The study protocol is approved by Human Ethical Committee (AUW/IHEC/BT-21-22/XPD-02) of Avinashilingam institute for Home Science and Higher Education for Women, Coimbatore, Tamil Nadu. (Annexure 2)

3.2.2 ADMINISTRATION OF QUESTIONNAIRE AND INFORMED CONSENT FORM

Each study subject received a questionnaire and was orally explained about the project and the questions asked in questionnaire. Before the sample collection the participants are signed in the informed consent form. (Appendix-1)

The study was designed with selected inclusion and exclusion criteria for the selection of participants

Inclusion criteria:

- Above 18 years
- Frequent smartphone users
- Neck pain for more than 15 days

Exclusion criteria:

- Alcoholic consumption, smoking habits
- Persons with diabetic, Cancer and other genetic disorder
- Persons with kidney, liver diseases, blood transfusion and any other pathological condition.

A prospective study observes for outcome, such as the development of disease, during study period and relates this to other factors such as suspected risk or protection factors.

Participants with known history of diabetes mellitus, kidney and liver diseases, blood transfusion and any other pathological conditions are excluded from the study because these known factors may obstruct the findings of outcome of the results. Participants who were less than eighteen years were not included due to ethical measures.

3.2.3 COLLECTION OF BLOOD SAMPLE

Peripheral venous blood samples (5ml) were collected from the subjects. All the samples are collected Vacutainers (AcCuvet-PLUS with clot activator) were centrifuged at 3000rpm at 4°C for 10 minutes. The samples were aliquoted into labelled cryovials and stored at -20°C for further analysis.

3.2.4 GROUPING OF PARTICIPANTS ON BASIS OF AGE

Based on the information collected from the participants they were further categorized into two groups as Group 1 (18-30) age Group 2 (31-45) age.

3.2.5 ASSESSMENT OF OXIDATIVE STRESS

Antioxidants are substances that can prevent or slow damage to cells caused by free radicals, unstable molecules that the body produces as a reaction to environmental and other pressures. They are sometimes called as free radical scavengers. Free radicals are waste substances produced as the body processes food and reacts to the environment. If the body cannot process and remove free radicals efficiently, oxidative stress can result. This can harm cells and body function. Free radicals are also known as reactive oxygen species (ROS).

3.2.5.1 Estimation of superoxide dismutase

Superoxide dismutase (SODs) are universal enzymes of organisms that live in the presence of oxygen. They catalyse the conversion of superoxide into oxygen and hydrogen peroxide. It speeds up certain chemical reaction in the body (Appendix 2).

3.2.5.2 Estimation of Glutathione peroxidase

Direct measurement of liberated free radical species is limited by their instability, the level of malondialdehyde (MDA), the stable product of oxidative degradation of polyunsaturated fatty acids, has been widely adopted as a measure of free radical formation. In previous study (Muukov *et al.* 2000), an increase of the serum MDA in reperfused patients with AMI was found shortly after successful thrombolytic treatment. The increase was followed by rapid transient stimulation of plasma GPx activity (Appendix 3).

3.2.5.3 Estimation of Reduced glutathione

Reduced glutathione is an antioxidant that protects the cells from damage caused by free radicals, heavy metals, and lipid peroxides. It can come as L-Glutathione reduced (GSH) or in an inactive state (GSSH). The ratio of reduced glutathione to oxidized glutathione within cells is a measure of cellular oxidative stress. In healthy cells and tissue, more than 90% of the total glutathione pool is in the reduced form (GSH), with the remainder in the disulfide form (GSSG) (Appendix 4).

3.2.5.4 Estimation of Vitamin A

Beta-carotene, another important lipid soluble membrane-bound antioxidant, is able to quench singlet oxygen interrupting the generation of ROS at a very early stage (Appendix 5).

3.2.5.5 Estimation of Vitamin E

Alpha-tocopherol is the major lipid soluble antioxidant which is present in the blood, mainly in the VLDL and LDL fractions, where it prevents free radicals to oxidize these lipoproteins. When incorporated into the membranes, α -tocopherol protects myocardial phospholipids (Gutteridge 1995) (Appendix 6).

3.2.5.6 Estimation of Vitamin C

Vitamin C in humans must be ingested for survival. Vitamin C is an electron donor, and this property accounts for all its known functions. As an electron donor, vitamin C is a potent water-soluble antioxidant in humans. Human diseases such as atherosclerosis and cancer might occur in part from oxidant damage to tissue (Appendix 7).

3.2.6 STATISTICAL ANALYSIS

SPSS package version 25.0 was used for the statistical analysis of data. SPSS (Statistical Package for the Social Sciences) is the set of software programs that are combined together in a single package. The basic application of this program is to analyse scientific data related with the social science. This data can be used for market research, survey, data mining, etc. The statistical level was set at $p < 0.05$. Independent T test and chi square test were performed for normally distributed data and if the data variation at significant level ($p < 0.05$).

Results and Discussion



RESULTS AND DISCUSSION

Mobile phone has become an integral part of modern communication in every individual. Over usage of technology leads to addiction. Currently, the addiction to smartphones among students is 24.8%–27.8%, and it is progressively increasing every year (Jeong, Lee., 2015). At the same time extensive usage of mobile phones leads to Self-reported symptoms associated with using mobile phones most commonly include headaches, earache, wrist pain and warmth sensations (Korpinen, Pääkkönen., 2009) and sometimes also perceived concentration difficulties and fatigue (Johansson *et al.*, 2010). However, EMF exposure due to mobile phone use is not currently known to have any major health effects. Researchers are studying tumour of the brain and central nervous system and other sites of the head and neck because mobile phones are typically held next to the head when used (National Cancer Institute, 2011)

Most smartphone tasks require users to stare sharply downwards and hold their arms out in front of them to see the screen, which makes the head move forward and causes an excessive anterior curve in the lower cervical vertebrae and an excessive posterior curve in the upper thoracic vertebrae to maintain balance (Kang *et al.*, 2012), (Berolo *et al.*, 2011), (Szeto *et al.*, 2002). This is known a head posture (FHP; turtle neck posture) and may impose exaggerated stresses on the cervical spine and neck muscles. Longer use was significantly associated with smartphone addiction, which is consistent with other studies that have found that increased exposure is linked with increased dependency (Cha *et al.*, 2019) Park & Lee (2012) also reported that smartphone use could be attributed to loneliness, depression and self-esteem based on their smartphone use and psychological well-being study. Mobile phones emit blue rays which on usage on low light can affect the human eye and has the high chance of having eye problem, eye irritation, sleeplessness, insomnia, redness in eyes, and may also cause myopia

The Lockdown scenario due to COVID' 19 has compelled people to adapt to the remote mode of working with the help of technology. Several literature studies have confirmed the adverse effects of excessive use of technology on human health (Antonio Moreno-Llamas *et al.*, 2020), (Deblina Roy *et al.*, 2020), (Auvinen *et al.*, 2019).

Younger people tend to use mobile phones higher than the old people. It has some working protocol where younger adult can easily adapt that, however still old people are trying learn the protocol to access the mobile phone. The biggest age-related difference lays in connectivity. Younger adults rated connectivity to computers and other devices and Internet access much higher than older adults (Cia Zhou., 2019).

Kesari *et al.* (2013) concluded that the mobile phone radiation and usage may increase the reactive oxygen species, which plays an important role in the development of metabolic and neurodegenerative diseases. Hence evaluation of oxidative biomarkers was conducted in the peripheral blood plasma of study volunteers to know the level of oxidative biomarkers in relation with the self-reported symptoms caused due to the extensive use of mobile phones.

The results of the present study are discussed under the following headings:

PHASE I

4.1. EXPLORATION OF IMPACT OF EXTENSIVE USAGE OF MOBILE PHONE THROUGH ONLINE QUESTIONNAIRE

- 4.1.1 Preparation of questionnaire based on over usage of mobile phones, its addiction and complication
- 4.1.2 Survey conducted through online among participants
- 4.1.3 Statistical Analysis
- 4.1.4 Demographic profile of the participants
- 4.1.5 Analysis of Habitual Behaviour of the participants
- 4.1.6 Addiction Behaviour of the participants
- 4.1.7 Awareness of mobile phone hazards among participants

PHASE II

4.2 EVALUATION OF OXIDATIVE MARKERS OF STUDY PARTICIPANTS

- 4.2.1 Estimation of superoxide dismutase

4.2.2 Estimation of Glutathione peroxidase

4.2.3 Estimation of Reduced glutathione

4.2.4 Estimation of Vitamin A

4.2.5 Estimation of Vitamin E

4.2.6 Estimation of Vitamin C

4.1 EXPLORATION OF IMPACT OF EXTENSIVE USAGE OF MOBILE PHONE THROUGH ONLINE QUESTIONNAIRE

4.1.1 Preparation of questionnaire based on over usage of mobile phones, its addiction and complication

Questionnaire was prepared and it was categorised under four topics namely demographic details of the participants which include the personal details of the participant like name, age, height, weight, educational qualification, accommodation. Habituation includes the questions like neck pain, occurrence of neck pain, sleeping position, degree of mobile phones usage, duration of sleeping time etc. Addiction behaviour complies the information regarding the addiction behaviour caused due to the extensive use of mobile phones like missing planned works, missing social life, feeling too much of time engaged on using mobile phone. And Awareness related questions namely EMR, neck pain occurrence, ear discomfort etc. were included.

4.1.2 Survey conducted through online among participants

Survey was conducted through online mode, where the questionnaire was distributed through social media. About 100 people of various age group participated in this online survey to maintain social distancing during the time of pandemic. The mode of conducting the survey was chosen to be online to maintain social distancing during the time of the Pandemic. A questionnaire was prepared in the form of Google Form which was composed of 36 questions self-designed by the investigators. The link of the questionnaire was forwarded to people email, WhatsApp, and Facebook to the contacts of investigators. Participants were encouraged to roll out the questionnaire to as many contacts as Table 1 includes specification of the participants like age, gender, height, weight, blood pressure, qualification, accommodation etc. Participants belonged to different ages ranging from 18 to 60 years having

work experience of 0–20 years were participated. To obtain a more precise analysis, age groups and work experience was divided into subgroups and small ranges. Moreover, the survey was limited to an educated class of people who know English, as, a questionnaire was framed in the English language. The following section describes and discusses about the information collected from the questionnaire and the statistical analysis. SPSS 25.0 was used for the analysis of data and chi square test was performed to know the significance of the data.

4.1.3 Statistical Analysis

SPSS- Statistical Package for the Social Sciences is a super easy software for analysing data and running statistical tests. It is versatile and responsive program designed to undertake a range of statistical procedures. Chi square test is used for the non-parametric analysis. It is used to examine between categorical variables from a random sample in order to judge goodness of fit between expected and observed results. Phi and Cramm's and Correlation was also used to find the significance among the participants.

4.1.4 Demographic profile of the participants

Table 1: Demographic profile of the participants

PARTICULAR	NUMBER OF PARTICIPANTS
Number of participants	100
• Male	28
• Female	72
Age group	
• 18-30	86
• 30-50	14

Academic Qualification	
• 10 th or 12 th / Diploma	5
• Undergraduate	55
• Postgraduate	39
• Doctorate	1
Occupation	
• Student	62
• Working	27
• Non-working	11
Accommodation	
• Hostel	32
• Home	68

Demographic details collect information about the personal details of the participants. It is found that from table 1 out of 100 participants 28% of them are male and 72% was female. Among 100 participants the age group is classified into 2 categories like age 18-30 as group 1 and age 31-50 and above as group 2, where 86% belongs to age group 1 which is considered as younger age and 14% belongs to age group 2 where they are considered as elder age group people. Academic profile of the participants was more like to be higher number in Under graduation. All of the participants are educated and so they could able to attend this survey. 5% of population completed 10th, 12 or diploma, 55% of population have done Under Graduation, 39% have completed Post Graduation and 1% doctorate have been found. Occupation includes more students of 62%, 27% of participants were working and 11% participants are non-working. The survey furthermore listed out the accommodation of the participants, where there is any change in the mobile phone usage in home and hostel. Here it is reported that 32% of participants are staying in hostel and 68% of participants are staying at home. It is that at hostel due to less restrictions people tend to use mobile phone widely.

4.1.5 Analysis of Habitual behaviour of the participants

Table 2: Habituation status of participants

HABITUATION	NO OF PARTICIPANTS	AGE GROUP 1	AGE GROUP 2	SIGNIFICANCE*
Type of mobile phone usage				
• Smart phone	97	84	13	0.012
• Basic phone	0	0	0	
• Both	3	2	1	
Total sleeping time				
• Below 5 hours	4	4	0	0.665
• 5-8 hours	79	67	12	
• Above 8 hours	17	15	2	
Sleeping position				
• Back	23	21	2	0.628
• Side	47	41	6	
• Foetal position	7	18	5	
• Stomach	23	6	1	
Number of years of mobile phone usage				
• 1-5 years	62	62	0	0.840
• 6-10 years	24	22	2	

• 11-15 years	6	1	5	
• Above 15 years	8	1	7	
Frequency of mobile phone checking				
• Rare	29	24	5	0.045
• Frequent	71	62	9	
Duration of mobile phone usage per day				
• Less than 1 hour	3	2	1	0.038
• 1-2 hour	20	15	5	
• 3-6 hour	51	46	5	
• More than 6 hours	26	23	3	
Posture of mobile usage				
• 0 degree	5	4	1	0.052
• 15degree	36	32	4	
• 30degree	31	24	7	
• 45degree	21	19	2	
• 60degree	7	7	0	

*Chi square test was performed ($p < 0.05$)

Habituation gives report of how participants are engaged with mobile phones and their usage. From table 2 we can see that most of them are using smartphones which has so many features where it makes people to engage with it for a long period of time. 97% participants were using smart phones and 3% were using both. Very smaller number of participants were having sleeping time less than five hours (4%) and that is age group 1. Younger generation like to use mobile phones largely at night time than adults. 67% of age group 1 is found to be having sleep of 5 to 8 hours a day. Participants of age group 2 (12%) seems to be having a sleeping hour of 5 to 8 hours a day. Very a smaller number of participants are having proper 8 hours of sleep a day. For sleeping position most of the participants prefer side 47%, back and stomach 23% and foetal 7 %. As most of the participants were students 1 to 5 years of mobile phone usage is higher (62%) and all are age group 1. 22% participants were using for about 6 to 10 years and 11-15 years (5%) above 15 (7%) were belongs to age group 2. Though age group 2 people have been using mobile phones for a longer period of time the impact of mobile phone technology is higher now due to the drastic development of the technology. It is also found that participants have the (71%) habit of checking mobile phone frequently where surprisingly both the age group 1(62%) and 2 (9%) have said that they have the habit of checking mobile phone. 29% of participants have the habit of checking mobile phones rare, which has resulted in the significance (0.045) with P value ($p < 0.05$). Very small number of participants have reported that they use mobile phones less than 1 hour. 20% are using 1-2 hours, 51% use mobile phones for 3 to 6 hours, where 46% belongs to age group 1 and 5% belongs to age group 2. 26% of participants are using mobile phones for more than 6 hours. Here along with age group 1 (23%) age group 2 (3%) are using mobile phones for more than 6 hours. Hence there is significance 0.038 as it is lesser than P value ($p < 0.05$). This shows the dependency of participants towards mobile phone extensively. Mobile phone posture includes 5% 0 degree, 36% 15 degree, 31% 30 degree, 21% 45degree and 7 % 60 degrees. It shows significance (0.052) as it is lesser than the p value ($p < 0.05$)

4.1.6 Addiction Behaviour of the participants

Addiction behaviour analysis shows the participants dependency towards the mobile phone. It is found that 52% of participants finding too much of time engaged in using mobile phones and 48% saying No. It shows significance 0.032 which is lesser than the P value $P < 0.05$. Minimum number of participants (38%) have said that missing planned work because of extensive usage of mobile phones.

Table 3: Addiction behaviour of participants

PARTICULARS	TOTAL NO	GROUP 1	GROUP 2	SIGNIFICANCE*
Finding too much time are engaged with mobile phone				
• Yes	52	47		0.032
• No	48	39		
Missing planned work because of mobile phone use	38	34	4	0.433
Yes	62	53	10	
No				
Using mobile phone causing any mental or psychological disturbance				
Yes	27	27	0	0.014
No	73	59	14	
Feeling of missing normal social life on usage of mobile phones	54	50	4	0.040
Yes	46	36	10	
No				

*Chi square test was performed ($p < 0.05$)

62% of participants reported that they are not missing planned works because of mobile phones. It is as well as found that out of 14%, 10% of age group 2 participants have said they are not missing any plans, which portray they are not dependent on mobile phones. Younger

people due to addiction behaviour they keep on scrolling social media or play games which occupy most of their time where they become lazy and schedule all the works later. This leads to missing of planned works higher in age group 1. Using mobile phones cause psychological disturbance shows significance of 0.014 which is lesser than $p < 0.05$, 27 % said yes and 73% said No. Group 2 age participants didn't have any psychological disturbance only younger people of age group 1 undergone psychological disturbance on extensive use of mobile phones. This could be due to not getting reply immediately, network issue, relationship problems they take it so seriously and may feel depression, anxiety, worried. Most of the younger participants of age group 1 (50%) now wish to spend time on mobile than spending time with family and friends, where majority of (10%) age group 2 have said that they don't feel like missing normal social like on usage of mobile phone. It also shows significance 0.040 which is lesser then the P value ($P < 0.05$).

4.1.7 Awareness of mobile phone hazards among participants

Table 4: Awareness analysis

PARTICULARS	TOTAL NO	AGE GROUP 1	AGE GROUP 2	SIGNIFICANCE*
Awareness about EMR				
• Yes	66	58	8	0.451
• No	34	28	6	
Having any wrist pain and hand pain because of smart phone use				
• Yes	35	27	8	0.049
• no	65	60	2	
Felt any discomfort while using mobile phone				
• Yes	28	34	4	0.023

• No	72	62	10	
Felt any ear discomfort while using mobile phone				
• Yes	22	21	1	0.148
• No	78	65	13	

**Chi square test was performed ($p < 0.05$)*

Awareness related question enquires about the pain, discomfort of the participants on extensive usage of mobile phone. From the questionnaire it is found that maximum number (66%) of population are aware of Electro Magnetic Radiation. Participants having wrist pain or hand pain is less in age group 1 where 8% of age group 3 reported that they undergone wrist and hand pain while using mobile phone. It shows significance value of 0.049 where it is lesser than $P < 0.05$. And moreover, participants feeling discomfort in very less number and this as well as shows significance value of 0.023 which is lesser than the p value $p < 0.05$. It is also found that participants do not have (78%) ear discomfort. Age group 2 (13%) haven't undergone any ear discomfort while using mobile phone. 21% of age group 1 have undergone ear discomfort on extensive use of mobile phone.

4.2 EVALUATION OF BIOMARKERS FROM STUDY VOLUNTEERS



Figure 2 Collection of blood sample from the participant



Figure 3 Blood sample before centrifugation

From the results of preliminary study 20 samples were collected from study volunteers. It is collected in a such a way 10 samples from age group 1 18-30 and 10 samples from age group 2 31-45. Blood sample (5.0 ml) (Figure 2,3) was collected and antioxidant assay have been estimated. The samples were correlated with the standard and regression graph was framed. The data were in the SPSS version 25.0 independent student t test is used for analysis and the results were summarized as follows;

4.2.1 Estimation Superoxide dismutase

Superoxide dismutase (SODs) constitute a very important antioxidant defence against oxidative stress in the body. The enzyme acts as a good therapeutical agent against reactive oxygen species.

Superoxide dismutase uses the photochemical reduction of riboflavin as oxygen generating systems and catalyses the inhibition of Nitro Blue Tetrazolium (NBT) reduction, the extent of which can be assayed spectrophotometrically at 600nm.

From independent sample test we could find that the significance value shows 0.038 where it is lesser than the p value $P < 0.05$. This shows that there is rise in the antioxidant due to the extensive usage of mobile phone

From figure 4 it is found that there is a significant change in the level of oxidative biomarkers, superoxide dismutase between two groups. Group 1 age people are more vulnerable on using mobile phones which can be clearly identified from the chart in figure 4. Whereas group 2 people show less change in the level of oxidative biomarker

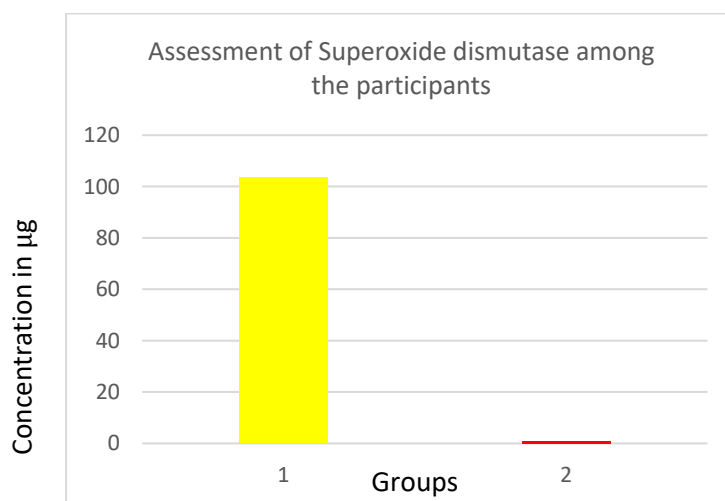


Figure 4: Comparison of Superoxide dismutase between two groups 1- Age group 1 (18-30), 2- Age group 2 (30-45)

4.2.2 Estimation of Glutathione peroxidase

Glutathione peroxidase is an antioxidant enzyme class with the capacity to scavenge free radicals. This in turn helps to prevent lipid peroxidation and maintain intracellular homeostasis as well as redox balance

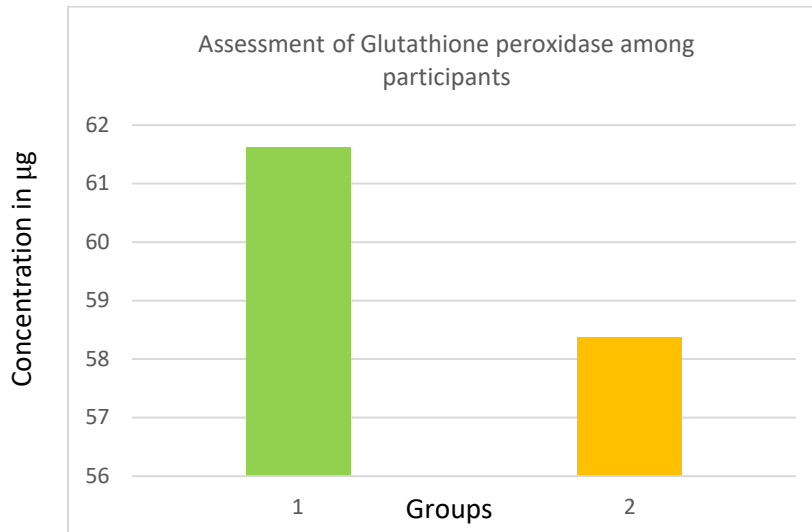


Figure 5: Comparison of Glutathione peroxidase between two groups, 1- Age group 1 (18-30), 2- Age group 2 (30-45)

Glutathione peroxidase shows greater significance value as it shows 0.001, so there may be some relation between the extensive use of mobile phones and rise in the level of glutathione peroxidase

Group 1 age have higher level of change in the oxidative biomarker than the group 2 age people (Figure 5).

4.2.3 Estimation of Reduced glutathione

Glutathione (GSH or reduced glutathione) is a tripeptide of gamma -Glutamyl cysteinyl glycine and the predominant intracellular organism including humans.

Reduced glutathione (GSH) is measured by its reaction with DTNB (5, 5-dithio- 2 - nitro benzoic acid) (Ellman's reaction) to give a compound that absorbs at 412nm.

From figure 6 we can identify that there is a huge difference in the level of oxidative biomarker. Both the age groups have identified having increase in the level of increase in the biomarker. But the level is higher in age group 1 people than the age group 2 people.

Reduced glutathione shows significance between the extensive usage of mobile phone. The significance value 0.37 is lesser than the probability value. So, there may be some relationship between the extensive mobile phone usage and increase in the level of reduced glutathione.

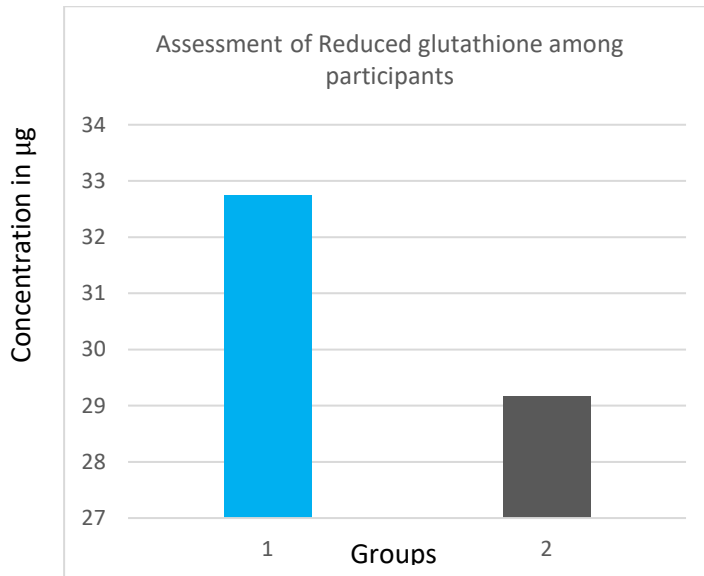


Figure 6: Comparison of Reduced Glutathione between two groups, 1- Age group 1 (18-30), 2- Age group 2 (30-45)

4.2.4 Estimation of Vitamin A

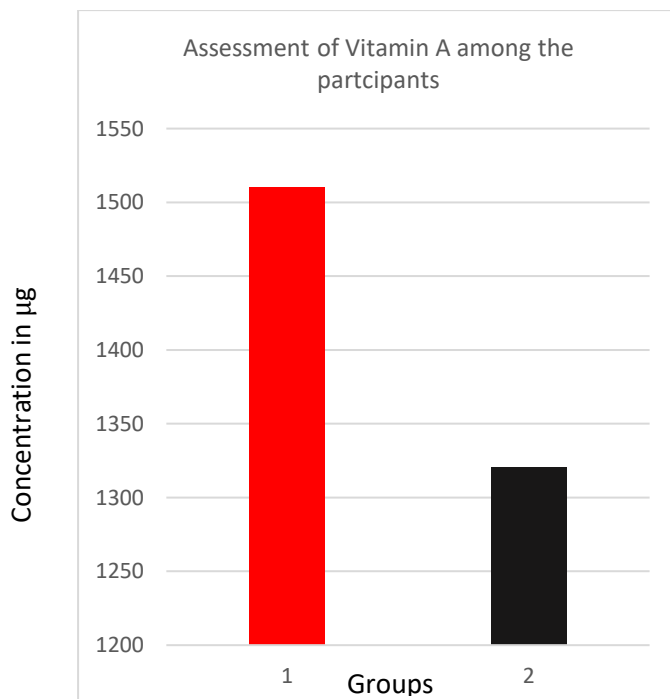


Figure 7: Comparison of Vitamin A between two groups, 1- Age group 1 (18-30), 2- Age group 2 (30-45)

Recent evidence has shown vitamin A, Carotenoids and pro vitamin A that carotenoids can be effective antioxidants for inhibiting the development of the heart disease. Vitamin A must be obtained from the diet: green and yellow vegetables, dairy products, fruits and organ meats are some of the richest sources.

The colour produced by vitamin A, its acetate or palmitate with TCA is proportional in intensity to its concentration, which is measured at 620 nm in a spectrophotometer.

Vitamin A doesn't show any significance as it shows 0.985 which says that there is no correlation between vitamin A and extensive usage of mobile phone.

Vitamin A is found to be increased in both of the age groups. From figure 7 we can spot that there is an increase in the level of oxidative biomarker in the age group 1 people than in age group 2 participants.

4.2.5 Estimation of Vitamin E

Vitamin E is a fat-soluble vitamin with several forms, but alpha- tocopherol is the only one used by the human body. Its main role is to act as an antioxidant, scavenging loose electrons -so called "free radicals" that damage cells. The Estimation follows (Quaife *et al.*, 1949).

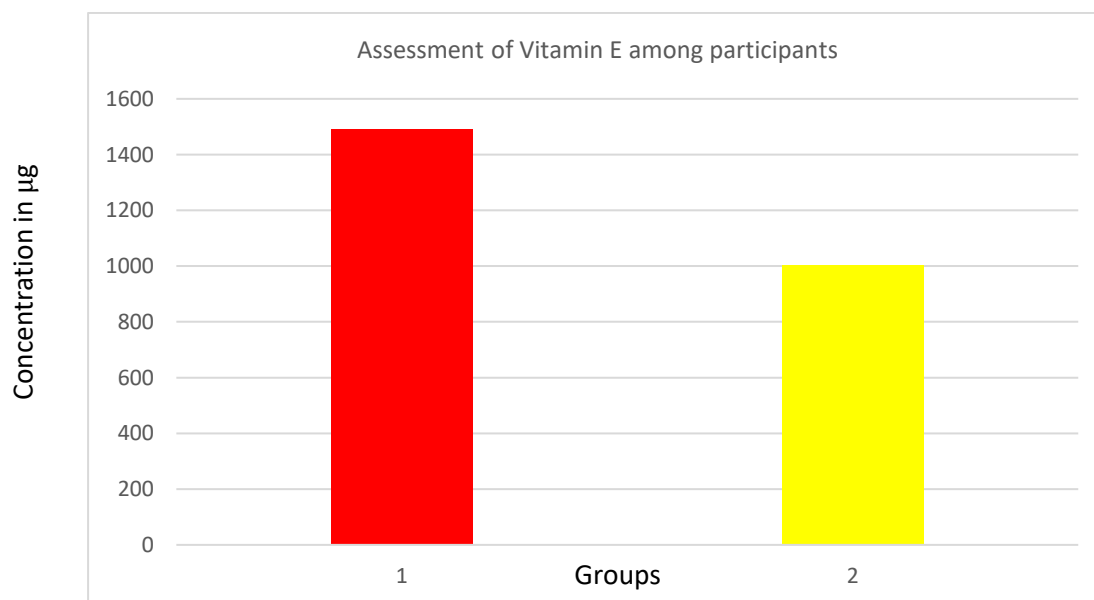


Figure 8: Comparison of Vitamin E between two groups, 1- Age group 1 (18-30), 2- Age group 2 (30-45)

Vitamin E can be estimated using Emmerie-Engel reaction which is based on the reduction of ferric to ferrous ions by tocopherols, which then forms a red colour with 2, 2'-

dipyridyl. Tocopherol and carotenes are first extracted with xylene and read at 460nm to measure carotenes. A correction is made for this after adding ferric chloride and read at 520nm.

In vitamin E the significance is 0.506 which is greater than the probability significance value. So, there is no significance here. This result that the level of antioxidant doesn't increase on over-usage of mobile phone.

From figure 8 it is noted that there is a raise in the level of oxidative marker in age group 1 people than in age group 2 people.

4.2.6 Estimation of Vitamin C

Vitamin C is also vital to your body's healing process. Vitamin C is an antioxidant that helps protect your cells against the effects of free radicals – molecules produced when the body breaks down food or exposed to tobacco smoke and the radiation from the solar radiation, X rays or other source

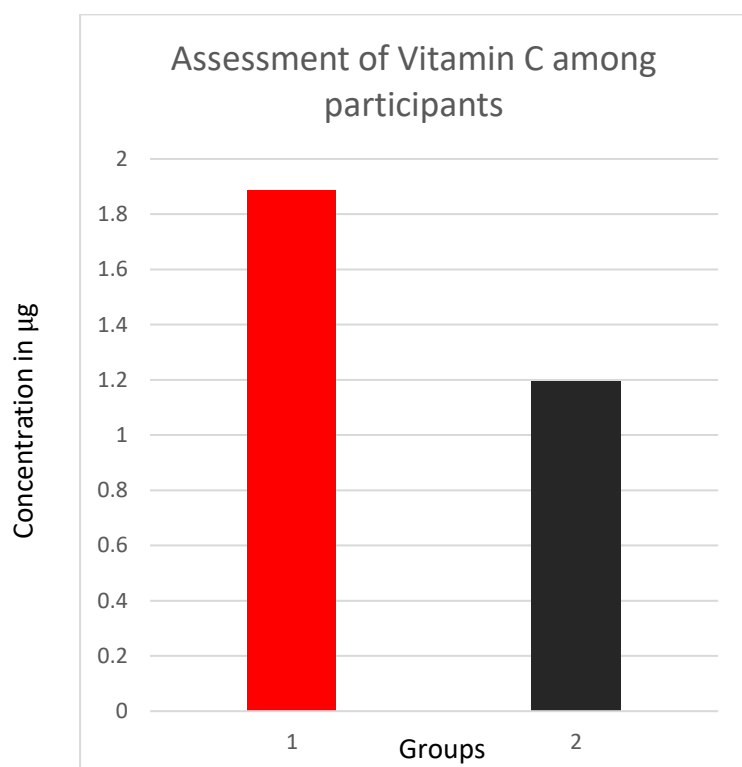


Figure 9: Comparison of Vitamin C between two groups, 1- Age group 1 (18-30), 2- Age group 2 (30-45)

Ascorbic acid is oxidised by copper to form dehydro-ascorbic acid and di-ketoglutaric acid. These products are treated with 2,4 dinitrophenyl hydrazine to form the derivative of bis 2,4 dinitro phenyl hydrazine. This compound in strong sulphuric acid undergoes a

rearrangement to form a product with an absorption band that is measured at 520 nm. The reaction is run in the presence of thiourea to provide a mildly reducing medium which helps to prevent interference from non-ascorbic acid chromogens

It is found that Vitamin C shows significance as it is 0.24 which is lesser than the P value $p < 0.05$, which states that there is a change in the level of biomarker.

From figure 9 it is clear that there is an increase in the level of biomarker in age group 1 people than in age group 2 people. But in both the age groups there is an increase in the level of vitamin C biomarker.

Among 100 participants 86 were younger age and 14 were old age group people. Both the age groups use smart phones. On the analysis of addiction behaviour younger age group are highly addicted towards the mobile phones. Both the age groups have the habit of checking mobile phones frequently which indicated that people are very much fond of using mobile phones. Younger age reported wrist and neck pain on extensive usage of mobile phone. Both the age groups (Group 1- 18-30, Group 2- 31-45) are using mobile phones more than 6 hours a day. Using mobile phone for a longer period on off light will affect the eyesight due to the blue light from the mobile phone. On addiction analysis younger people are found to be more active with mobile phones all the time, which results in missing of social life, delay in work progress, laziness etc

According to the finding of phase I, they were grouped into two (Group I- 18-30, Group II 39-45), it is found that both the age groups undergo certain stress which resulted in increase in the level of oxidative biomarker. Enzymatic biomarkers like Superoxide dismutase, glutathione peroxidase, reduced glutathione was found to be increased in both the age groups. Elder people are less affected than the younger people. There is a significant increase in superoxide dismutase biomarker level in younger people and in elder people the stress is found to be less. Non enzymatic biomarkers like Vitamin A, Vitamin C, Vitamin also showed some level of increase in the biomarkers. However younger group have the highest level of oxidative marker than the elder people. Both the age groups are using mobile phones, yet younger people relay on mobile phone all the time. As well as elder people are using mobile phone but comparatively less than younger group, maybe they could not able to understand the features and technology to operate mobile phones as younger people.

Summary and Conclusion

SUMMARY AND CONCLUSION

The use of mobile phones has become extensively popular in recent years. Despite the many benefits that mobile phones have as an opportunity, excessive use of them can cause various problems such as mobile addiction. Mobile phone addiction stimulates urge in people to use extensively for a longer period of time. It leads to many external and also internal problems like eye irritation, laziness, insomnia, tiredness, wrist pain, neck pain etc.

Now mobile phone has become an integral part of the body. All the age group people are now fond of mobile phones. Extensive use of mobile phone creates additional stress to the body. Oxidative stress leads to change in the level of oxidative biomarkers

Hence it is imperative to conduct the evaluation of oxidative biomarkers in relation with the self-reported symptoms among extensive usage of smartphone users on the basis of age which may help to examine age-related differences in requirements for mobile phones in the context of the transition from feature phones to smart phones

Objectives of the study

- To explore the impact of extensive usage of smartphone users through online questionnaire.
- To evaluate oxidative biomarkers in the blood sample of over-usage of smartphone users.
- To determine the levels of oxidative marker related to age among extensive usage of smartphone users.

The study was conducted in two phases

PHASE I

Exploration of impact of extensive usage of mobile phone through online questionnaire

Online survey was conducted. A total number of 100 people participated in the survey. This survey includes the collection of information like demographic details of the participants, Analysis of habitual behaviour of the participants, awareness of mobile phone hazards by the participants and addiction behaviour of the participants on over usage of mobile phones. These questions carry the information about the personal details of the participants like age, gender,

height, weight, occupation, academic qualification, pain caused by the over usage of mobile phones, their addiction towards the mobile phones.

Salient findings

- Among 100 participants 86 were younger age and 14 were old age group people. Both the age groups use smart phones.
- On the analysis of addiction behaviour younger age group are highly addicted towards the mobile phones
- Both the age groups have the habit of checking mobile phones frequently which indicated that people are very much fond of using mobile phones.
- Younger age reported wrist and neck pain on extensive usage of mobile phone.
- Both the age groups (Group 1- 18-30, Group 2- 31-45) are using mobile phones more than 6 hours a day. Using mobile phone for a longer period on off light will affect the eyesight due to the blue light from the mobile phone.
- On addiction analysis younger people are found to be more active with mobile phones all the time, which results in missing of social life, delay in work progress, laziness etc.

PHASE II

Evaluation of oxidative markers

According to the self-reported symptoms of participants who have complication on extensive usage of mobile phone, 20 participants were selected for the evaluation of oxidative markers. Questionnaire and informed concern were signed and 5 ml of blood sample was collected from the participants. Evaluation of Oxidative biomarkers like enzymatic includes Superoxide dismutase, Glutathione peroxidase reduced glutathione and non-enzymatic includes Vitamin A, Vitamin C, Vitamin E to know the level of oxidative biomarker were performed.

Salient findings

- According to the finding of phase I, they were grouped into two (Group I- 18-30, Group II 39-45), it is found that both the age groups undergo certain stress which resulted in increase in the level of oxidative biomarker
- Enzymatic biomarkers like Superoxide dismutase, glutathione peroxidase, reduced glutathione was found to be increased in both the age groups. Elder people are less

affected than the younger people. There is a significant increase in superoxide dismutase biomarker level in younger people and in elder people the stress is found to be less

- Non enzymatic biomarkers like Vitamin A, Vitamin C, Vitamin also showed some level of increase in the biomarkers. However younger group have the highest level of oxidative markers than the elder people.
- Both age groups are using mobile phones, yet younger people rely on mobile phone all the time. As well as elder people are using mobile phone but comparatively less than younger group, maybe they could not able to understand the features and technology to operate mobile phones as younger people.

Conclusions drawn from the findings of the present investigation

Pain and stress may cause increase in the level of oxidative biomarkers occurred due to extensive usage of mobile phones found from the self-reported symptoms from the participants.

Evaluation of biomarkers from participants resulted in increase in the level of oxidative biomarkers. Both the age groups found to have increased level of biomarkers but younger group found have higher change in oxidative markers than the elder group.

The study clearly suggested that an extensive usage of mobile phones may cause many changes in the metabolism that leads to the generation of ROS, which results in increase in the level of oxidative biomarker.

Scope for the future studies

- ✓ A study can be conducted on whether the increase in the level of oxidative biomarkers leads to any other problem like tumour, cancer, musculoskeletal problem or any other issues related to the pain
- ✓ Some more biomarkers can also be included to find the level of increase in the biomarkers caused due to the extensive usage of mobile phones

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Appendix



APPENDICES

Appendix I

Questionnaire Administered to the Study Volunteers

1. Name:
2. Age:
3. Gender:
4. Height:
5. Weight:
6. Blood group:
7. Blood pressure level:
80-120/ Above 140/ Below 80
8. Academic qualification:
10th or 12th/ diploma/ Undergraduate/ Postgraduate/ Doctorate/ None
9. Occupation:
Student/ Working/ Nonworking
10. Accommodation:
Home/ Hostel
11. Mobile phone user:
Yes/no
12. Type of mobile phone usage:
Smart phone/ Basic phone/ Both
13. Have undergone any neck pain on over usage of mobile phone:
Yes/ No/ may be
14. Occurrence of neck pain:
Rare/ frequent
15. Have undergone any major or minor accident or surgery:
Yes/ No
16. If yes, specify
17. Total sleeping time:

Below 5 hours/ 5-8 hours/ above 8 hours

18. Sleeping Position:

Back/ Side/ Foetal position/ Stomach

19. Number of years of mobile phone usage:

1-5 years/ 6-10 years/ 11-15 years/ above 15 years

20. Duration of mobile phone usage per day:

Less than 1 hour/ 1-2 hrs/ 3-6 hrs/ More than 6 hrs

21. Frequency of mobile phone checking:

Frequent/ Rare

22. Checking of mobile phone in between sleep:

Yes/ No/ Rare

23. Felt any discomfort while using mobile phone:

Yes/ No

24. Felt any ear discomfort while using mobile phone:

Yes/ No

25. Place of mobile phone keeping:

In the bag/ Around your pelvic area/ Around your neck

26. Most frequently used app:

Entertainment/ Photography/ Communication/ Education/ Game/ social media/
Reading books/ Job related/ Business/ Others

27. Have you aware of EMR:

Yes/ No

28. Having any pain on wrist and hand because of smart phone use:

Yes/ No

29. Any medication for neck pain:

own medication/ Doctor prescribed / none

30. Preference of medication:

Tablets/ Ointment/ Spray/ Physical therapy/ None

31. Any other complication on over usage of mobile phone along with neck pain?

32. Posture of mobile phone usage:

0 degree/15 degree/30 degree/ 45 degree/ 60 degrees

33. Finding too much of time are engaged with mobile phone:

Yes/ No

34. Missing planned work because of mobile phone use:

Yes/ No

35. Using of mobile causing any mental or psychological disturbance:

Yes/ No

36. Feeling of missing normal social life on usage of mobile phone:

Yes/ No

Informed consent

Informed consent form for study volunteers who are participating in the research on evaluation of oxidative biomarkers on extensive usage of mobile phones

The title of the research project is “Evaluation of oxidative biomarkers in relation with the self-reported symptoms among on extensive usage of mobile phones

Principal Investigator : **Shirley A**

Organisation : **Department of Biochemistry, Biotechnology and Bioinformatics, Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore- 641 043, Tamil Nadu, India.**

Name of the proposal : **Evaluation of oxidative biomarkers in relation with the self-reported symptoms among extensive usage of smartphone users on the basis of Age**

The informed consent form has two parts:

1. Information sheet (to share information about the research with you)
2. Certification of consent (for signature if you agree to take part)

You will be given a copy of the full Informed consent form

Part I: Information sheet

Introduction

I am A. Shirley, Student, Department of Biochemistry, Biotechnology and Bioinformatics, Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore. I am carrying out a study on the topic of Evaluation of Oxidative Biomarkers in relation with the self-reported symptoms among extensive usage of smart phone users on the basis of age as part of my research project. Before you decide, you can talk to anyone you feel

comfortable with about the research. There may be some words that you do not understand. Please ask me to explain. If you have any questions later, you can ask the staff or me.

Purpose of the research

Mobile phone over-usage leads to clinical complications in our body, in such condition where the body produce ROS response which in turn increase the oxidative biomarker in our body. This study is being done to associate the level of oxidative biomarkers from the blood sample collected from mobile phone over users. Also, to find the specific oxidative marks which expressed in higher level and its role in creation of stress.

Type of Intervention

This research will involve in collection of blood samples to determine the change in the oxidative biomarkers on extensive usage of mobile phone.

Selection of the participants

We are inviting all adults in the age group of 18-45 who use mobile phones in their day today life and undergone any physiological stress like neck pain, wrist discomfort in and around Coimbatore, to participate in the evaluation of oxidative biomarkers.

1. Do you know why we asking to take part in this study?
2. Do you know what the study about?

Voluntary participation

Your participation in this research is entirely voluntary. It is your choice whether to participate or not. You may change your mind later and stop participating even if you agreed earlier.

1. Do you know that you do not have to take part in this research study, if you do not wish to?
2. Do you have any questions?

Procedure and protocol

A. Unfamiliar procedures

We will take blood from your arm using syringe and needle. We will take about 5ml of blood. At the end of the research, in 1 year, any leftover blood samples will be destroyed.

B. Description of the process

1. At first, we will ask you to answer the questions we have and fill up a questionnaire, which asks for personal and personal details.
2. Then, about 5.0 ml of blood, equal to about a teaspoon, will be taken from your arm with a syringe. This blood will be tested for the identification of change in the oxidative biomarker
 - a. We will also ask you a few questions about your general health and measure how tall you are and how much your weight

Duration

The research takes place over a period of 6 months.

1. Can you tell me if you remember, the number of times that we are asking you to come to the hospital to complete the treatment?
2. The research projects?
3. How much blood will be taken from your veins, using a syringe and needles?
4. Do you have any other questions?
5. Do you want me to go through the procedures again?

Side Effects

There are no potential side effects dur to this study

Risks

There is no potential risk in participating in this study

Benefits

There may not be any benefits but it is likely to help us to find change in oxidative biomarkers on over usage of smart phone users. There may not be any benefits to the society at this stage of research, but in future we may be able to tell whether on extensive usage of mobile phone causes any change in the level of oxidative biomarker and to bring awareness to society.

Reimbursements

You will not be given money or gifts to take part in this research.

1. Can you tell me if you have understood correctly the benefits that you will have if you take part in the study?

2. Do you know if the study will pay for your travel costs?
3. Do you have any other questions?

Confidentiality

With this research, something out of the ordinary is being done in your community. It is possible that if others in the community are aware that you are participating, they may ask you questions. We will be sharing the identity of those participating in the research. The information that we collected from this research project will be kept confidential. The information about you that will be collected during the research will be put away and no one but the researchers will be able to see it. Any information will have a number on it instead of your name. Only the researchers will know what the number is and we lock that information with lock and key. It will not be shared with or given to anyone.

1. Did you understand the procedures that we will be using to make sure that any information that we as researchers collect about you will remain confidential?
2. Did you have any question about this?

Sharing the results

We plan to give you the result of level of antioxidants in your blood.

Right to refuse or withdraw

You do not have to take part in this research if you do not wish to do so. You may also stop participating in the research at any time you choose. It is your choice and all of your rights will still be respected.

Whom to contact

If you have any questions, you ask them now or later, even after the study has started. If you wish to ask question later, you may contact any of the following

A. Shirley, Student, Post Graduate Student, Department of Biochemistry, Biotechnology and Bioinformatics, Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore- 641043, Ph: 8608354316, Email: shirley.anthony.9212@gmail.com

This proposal has been reviewed and approved by Institutional Human Ethics Committee, (AUW/IHEC/BT-21-22/XPD-02) Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore- 641043, which is a committee whose task is to

make sure that research participants are protected from harm. If you wish to find more about the Institutional Review Board, contact,

Dr. S. Uma Mageshwari, Member Secretary, Institutional Human Ethics Committee, Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore-641043

Part II: Certificate of Consent

The above information regarding the study, has been read by me/ read to me, and has been explained to me by the investigator(s). Having understood the same, I hereby give my consent to them to interview me, and collect biological sample 5ml Blood from me. I am affixing my signature / left thumb impression to indicate my consent and willingness to participate in this study (i.e., willingly abide by the project requirements)

Signature / Left thumb impression of the Study Volunteer / Legal Representative:

Signature of the Interviewer with date: _____

Signature of the Witness with name: _____

Statement by the Researcher/Person taking Consent

I have accurately read out the information sheet to the potential participant and to the best of my ability made sure that the participant understands that the following will be done:

1. Collection of 5 ml of blood sample

I confirm that the participant was given an opportunity to ask questions about the study and all the questions asked by the participants have been answered correctly and to the best of my ability. I confirm that the individual has been coerced into given consent and the consent has been freely and voluntarily.

A copy of this ICF been provided to the participant

Name of the Researcher/ Person taking the consent: _____

Signature of Researcher/ Person taking the consent: _____

Date: _____

Appendix-2
Estimation of Superoxide dismutase activity
Spectrophotometry method
(Beauchamp and Fridovich, 1971)

Principle

Superoxide dismutase uses the photochemical reduction of riboflavin as oxygen generating systems and catalyses the inhibition of Nitro Blue Tetrazolium (NBT) reduction, the extent of which can be assayed spectrophotometrically at 600nm.

Reagents

1. 250mM Potassium phosphate buffer (pH 7.8)
2. 100mM Methionine
3. 10mM Riboflavin
4. 750 μ M Nitro Blue Tetrazolium (NBT) salt
5. 5mM EDTA

Procedure

Mixed a 3ml reaction cocktail containing: 50mM potassium phosphate buffer, 13mM methionine, 2 μ M riboflavin, 0.1mM EDTA and 75 μ l NBT. To the test 0.1 ml of plasma was added. The reagents were added in dark. Reduced NBT was measured spectrophotometrically at 560nm after exposure to light for 15 minutes. The 50% inhibition of the reaction between riboflavin and NBT in the presence of methionine is taken as 1 unit of SOD activity. The enzyme activity was expressed as units/ml of sample.

Pipetted into tubes marked	Blank	Standard	Test
Reagents			
Methionine(ml)	2.5	2.5	2.5
Riboflavin(ml)	0.3	0.3	0.3
NBT (ml)		0.1	0.1
0.4M Phosphate buffer (ml)	0.2	0.1	
Plasma (ml)			0.1

Appendix-3

Estimation of Glutathione peroxidase activity

Spectrophotometry method

(Rotruck *et al.*, 1973)

Reagents:

1. 0.4M Sodium phosphate buffer (pH 7.0)
2. 10mM Sodium azide
3. 4mM Reduced glutathione
4. 2.5mM H₂O₂
5. 10% Trichloro acetic acid
6. Phosphate solution: 0.3mM Disodium hydrogen phosphate
7. DTNB reagent: 40 mg of 5, 5 dithiobis (2-nitro benzoic acid) / 100ml of 1% Sodium citrate
8. Reduced glutathione standard: 20 mg percent solution.

Procedure:

0.4 ml of buffer, 0.1 ml sodium azide, 0.2 ml reduced glutathione, 0.1 ml of plasma, 0.1 ml hydrogen peroxide and water were taken to a final incubation volume of 2.0 ml. The

test tubes were incubated at 30°C for 10 minutes. The reaction was terminated by the addition of 0.5 ml of trichloroacetic acid. To determine the residual GSH content, 1 ml supernatant was removed by centrifugation and added 3ml of disodium hydrogen phosphate and 1ml of DTNB reagent. The colour was read at 412 nm. A blank was prepared with only disodium hydrogen phosphate and 1 ml of DTNB reagent. Suitable aliquots of the standard were taken and treated similarly. The activity was expressed as µg of glutathione consumed/minute/ml of sample.

	Conc	Buffer(ml)	Sodium azide (ml)	Reduced Glutathione (ml)	Hydrogen peroxide (ml)	H ₂ O (ml)	Incubate at 37°C for 10 mins	TCA (ml)	Centrifuged for 10mins	Supernatant (ml)	Disodium hydrogen phosphate (ml)	DTNB (ml)	Optical density 412 nm
Blank										1			
Standard S1	0.1	0.4	0.1	0.2	0.1	1.1		0.5		1	3	1	
S2	0.2	0.4	0.1	0.2	0.1	1		0.5		1	3	1	
S3	0.3	0.4	0.1	0.2	0.1	0.9		0.5		1	3	1	
S4	0.4	0.4	0.1	0.2	0.1	0.8		0.5		1	3	1	
S5	0.5	0.4	0.1	0.2	0.1	0.7		0.5		1	3	1	
Test	0.1	0.4	0.1	0.2	0.1	1.1		0.5		1	3	1	

Appendix-4

Estimation of Reduced glutathione

Spectrophotometry method

(Moron *et al.*, 1979)

Principle

Reduced glutathione (GSH) is measured by its reaction with DTNB (5, 5-dithio- 2 - nitro benzoic acid) (Ellman's reaction) to give a compound that absorbs at 412nm.

Reagents

1. DTNB solution: 0.6mM 5, 5-dithio- 2 -nitro benzoic acid in 0.2M phosphate buffer.

2. 5% Trichloroacetic acid
3. 0.2M Sodium phosphate buffer (pH 8.0)
4. Glutathione standard: 10mg Reduced glutathione in 100ml of 5% TCA

Procedure

0.1 ml of plasma and 0.2 ml of trichloroacetic acid were added and centrifuged for 15 minutes at 1500 rpm. 2.0 ml of dithionitrobenzoic acid (DTNB) was added to 0.1 ml of the supernatant and the total volume made up to 3.0 ml with 0.2M phosphate buffer. The colour developed was read at 412 nm in a UV visible spectrophotometer.

	Conc	5% TCA (ml)	Centrifuge 15mins At 1500 rpm	Supernatant (ml)	Dithionitrobenzoic Acid (ml)	0.2M Phosphate buffer	Optical density(412 nm)
Blank							
Standard S1	0.2	0.2		0.1		0.9	
S2	0.4	0.2		0.1		0.9	
S3	0.6	0.2		0.1		0.9	
S4	0.8	0.2		0.1		0.9	
S5	1.0	0.2		0.1		0.9	
Test	0.1	0.2		0.1		0.9	

Appendix-5

Estimation of Vitamin A

Spectrophotometry method

(Bayfield and Cole, 1980)

Principle

The colour produced by vitamin A, its acetate or palmitate with TCA is proportional in intensity to its concentration, which is measured at 620 nm in a spectrophotometer.

Reagents

All reagents were prepared fresh. Exposure of samples and reagents to light was avoided at all times.

1. Saturated Trichloro acetic acid (TCA) in chloroform: 15 g clear TCA crystals were dissolved in 25 ml of alcohol- free chloroform, stored in dark.
2. Standard vitamin A: 1.5 mg vitamin A palmitate / 10 ml of chloroform.
3. Petroleum ether (40-60°C)
4. 95% ethanol

Vitamin A extraction

Equal volume of ethanol was added to 0.1 ml plasma and shaken with 0.3 ml petroleum ether for 3minutes. This was centrifuged to separate the layers. Petroleum ether layer was pipetted, evaporated to dryness, dissolved in 0.5ml alcohol- free chloroform and was taken as test.

Procedure

Aliquots of the standard were pipetted out into a series of clean, dry test tubes and 0.2 ml test was taken in another test tube. The volumes in all the tubes were made up to 1.0 ml with chloroform. 2 ml of TCA reagent was added and rapidly mixed with the contents of the tube. The absorbance was recorded immediately at 620 nm in a spectrophotometer. Constructed a standard graph and read off the concentration in the sample.

	Conc(ml)	Chloroform(ml)	TCA (ml)	Rapidly mixed With Content immediately	Optical density (620nm)
Blank					
Standard (s1)	0.2	0.8	2		
S2	0.4	0.6	2		
S3	0.6	0.4	2		
S4	0.8	0.2	2		
S5	1.0	0	2		

Test	0.2	0.8	2		
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Appendix-6
Estimation of Vitamin E
Spectrophotometry method
(Quaife *et al.*, 1949)

Principle

Vitamin E can be estimated using Emmerie-Engel reaction which is based on the reduction of ferric to ferrous ions by tocopherols, which then forms a red colour with 2, 2'-dipyridyl. Tocopherol and carotenes are first extracted with xylene and read at 460nm to measure carotenes. A correction is made for this after adding ferric chloride and read at 520nm.

Reagents

1. Absolute alcohol
2. Xylene
3. 2, 2'-dipyridyl: 1.2 g in 1 litre of N- propanol.
4. Ferric chloride solution: 1.2 g of $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ in 1 litre of ethanol and kept in a brown bottle.
5. Standard solution: Dissolved 100mg of tocopherol acetate in 1liter absolute alcohol.

Procedure

Into 3 stoppered centrifuge tubes (test, standard and blank) pipetted out 0.3 ml of plasma, 0.3 ml of standard and 0.3 ml of water respectively. Added 1.0 ml ethanol, 1.0 ml xylene to all the test tubes, stoppered, mixed well and centrifuged. Transferred 1.0 ml of xylene layer into another stoppered tube, taking care not to include ethanol layer. Added 1.0 ml of 2, 2'-dipyridyl reagent to each tube, stoppered and mixed. The test and standard were read against the blank at 460nm. Then beginning with the blank, in turn, added 0.33 ml of ferric chloride solution. After 15 minutes the test and standard were read against the blank at 520 nm.

		Ethanol	Xylene	Centrifuged for 10mins To separate the xylene layer	Supernatant(ml)	2,2-Dipyridyl (ml)	Optical density (460nm)	Chloride(ml)	Optical density(520nm)
Blank	0.3	1	1		1	0.1		0.33	
Standard	0.3	1	1		1	0.1		0.33	
Test	0.3	1	1		1	0.1		0.33	

Appendix – 7

Estimation of Vitamin C

Spectrophotometry method

(Omaye *et al.*, 1979)

Principle

Ascorbic acid is oxidised by copper to form dehydroascorbic acid and Di ketoglutaric acid. These products are treated with 2,4 dinitrophenyl hydrazine to form the derivative of bis 2,4 dinitro phenyl hydrazine. This compound in strong sulphuric acid undergoes a rearrangement to form a product with an absorption band that is measured at 520 nm. The reaction is run in the presence of thiourea to provide a mildly reducing medium which helps to prevent interference from non-ascorbic acid chromogens.

Reagents

1. 5% TCA
2. 65% Sulphuric acid
3. DTCS reagent: 3 g of 2,4 DNPH, 0.4 g thiourea and 0.05 copper sulphate dissolved in 9 N sulphuric acid and made up to 100 ml.
- 4 Standard: Ascorbic acid in the range of 4-20 µg/ml was prepared in 5% oxalic acid.

Procedure

0.1 ml of plasma was precipitated with 5% ice-cold TCA and centrifuged for 20 min. 0.1 ml of the supernatant was mixed with 0.02 ml of DTCS reagent and incubated for 3 hours

at 37°C. Then 0.15 ml of ice-cold 65% sulphuric acid was added. Mixed well and the solutions were allowed to stand at room temperature for an additional 30 min. Absorbance was determined at 520 nm. The results were expressed as mg/dl.

	Conc (ml)	Oxalic acid (ml)	DTCS (ml)		65% H ₂ SO ₄ (ml)	Mixed well & Additional 30 mins in room temperature	Optical density (520nm)
Blank				Incubate At 37°C For 3 hours			
Standard S1	0.2	0.8	0.02		0.15		
S2	0.4	0.6	0.02		0.15		
S3	0.6	0.4	0.02		0.15		
S4	0.8	0.2	0.02		0.15		
S5	1.0	0	0.02		0.15		
Test	0.1	0.9	0.02		0.15		

Annexure



ANNEXURE

Annexure I

Online questionnaire

<p>Analysis of complication on over usage of mobile phone</p> <p>A survey of results on mobile phone over usage based on age</p> <p>Email *</p> <p>Valid email</p> <p>This form is collecting emails. Change settings</p>	<p>Height *</p> <p>Short answer text</p>
<p>Name (Capital) *</p> <p>Short answer text</p>	<p>Weight *</p> <p>Short answer text</p>
<p>Age *</p> <p><input type="radio"/> 18-30</p> <p><input type="radio"/> 30-45</p> <p><input type="radio"/> Above 45</p>	<p>Blood Group *</p> <p>Short answer text</p>
<p>Gender *</p> <p><input type="radio"/> Male</p> <p><input type="radio"/> Female</p>	<p>Blood pressure level *</p> <p><input type="radio"/> 80-120</p> <p><input type="radio"/> Above 140</p> <p><input type="radio"/> Below 80</p>
<p>Occupation *</p> <p><input type="radio"/> Student</p> <p><input type="radio"/> Working</p> <p><input type="radio"/> Non working</p> <p><input type="radio"/> Other...</p>	<p>Academic Qualification *</p> <p><input type="radio"/> 10th or 12th / diploma</p> <p><input type="radio"/> Undergraduate</p> <p><input type="radio"/> Postgraduate</p>
<p>Accommodation *</p> <p><input type="radio"/> Home</p> <p><input type="radio"/> Hostel</p>	<p>Have undergone any neck pain on over usage of mobile phone? *</p> <p><input type="radio"/> Yes</p> <p><input type="radio"/> No</p> <p><input type="radio"/> May be</p>
<p>Mobile phone users *</p> <p><input type="radio"/> yes</p> <p><input type="radio"/> no</p>	<p>Occurrence of neck pain *</p> <p><input type="radio"/> Rare</p> <p><input type="radio"/> Frequent</p>
<p>Type of mobile phone usage *</p> <p><input type="radio"/> Smart phone</p> <p><input type="radio"/> Basic phone</p> <p><input type="radio"/> Both</p>	<p>Have undergone any major or minor accident or surgery *</p> <p><input type="radio"/> yes</p> <p><input type="radio"/> no</p>
	<p>... If yes, specify</p> <p>Short answer text</p>
	<p>Total sleeping time *</p> <p><input type="radio"/> Below 5 hours</p> <p><input type="radio"/> 5 - 8 hours</p>

Sleeping position *



- Back
- Side
- Fetal position
- Stomach

Number of years of mobile phone usage * :::

- 1 - 5 years
- 6 - 10 years
- 11 - 15 years
- Above 15 years

Duration of mobile phone usage per day *

- Less than 1 hour
- 1 - 2 hours
- 3 - 6 hours
- More than 6 hours

Frequency of mobile phone checking *

- Frequent
- Rare

Checking of mobile phone in between sleep :::

- Yes
- No
- Rare

Felt any discomfort while using mobile phone *

- Yes
- No

Felt any ear discomfort while using mobile phone *

- Yes
- No

Have you aware of Electromagnetic radiation ? *

- Yes
- No

Place of mobile phone keeping *

- In the bag
- Around your pelvic area (pocket)
- Around your neck

Having any pain on wrist & hand because of smart phone use ? *

- Yes
- No

Most frequently used application * :::

- Entertainment
- Photography
- Communication
- Education
- Game
- Social media
- Reading books
- Other...

Any medication for neck pain ? * :::

- Own medication
- Doctor prescribed medicine
- None

Preference of medication ? *

- Tablets
- Ointment
- Spray
- Physical therapy
- None

Posture of mobile usage ? *



- 0 degree
- 15 degree
- 30 degree
- 45 degree
- 60 degree

Finding too much of time are engaged with mobile phone ? *

- Yes
- No

Missing planned work because of mobile phone use ? *

- Yes
- No

Using of mobile causing any mental or psychological disturbance ? *



- Yes
- No

Feeling of missing normal social life on usage of mobile phone? *

- Yes
- No

Annexure 2

Ethical approval for all studies involving Human participants

INSTITUTIONAL HUMAN ETHICS COMMITTEE  Avinashilingam Institute for Home Science and Higher Education for Women (Deemed to be University under Category 'A' by MHRD, Estd. u/s 3 of UGC Act 1956) Re-accredited with 'A++' Grade by NAAC. Recognised by UGC Under Section 12 B Coimbatore-641 043, Tamil Nadu, India	
<p>Chairman Dr. SudhaRamalingam Director-Research & Innovation, Professor-Community Medicine, PSG Institute of Medical Sciences & Research, Coimbatore</p> <p>Member Secretary Dr.S.Uma Mageshwari Professor and Head, Department of Food Service Management & Dietetics</p> <p>Members Mr.K.Arunmoli (Legal Expert) Dr.Subhashini K. Sripathi Dr.A.Saraswathy (Medical Officer) Ms.D.Kavitha Dr.A.R.SudamaniRamasamy Dr.G.Victoria Naomi Dr. Judith Justin Dr.AnithaSubash</p>	<p style="text-align: right;">22nd Febraury 2022</p> <p>To Ms.A.Shirley Department of Biotechnology Avinashilingam Institute for Home Science and Higher Education for Women Coimbatore – 641 043</p> <p>Dear A.Shirley, Ref: Your proposal No. IHEC/21-22/BT-02 entitled “Evaluation of Oxidative Biomarkers in Relation with the Self- reported Symptoms among over usage of Smart Phone users on the basis of age” submitted for approval of IHEC on 23.11.2021</p> <p>The Institutional Human Ethics Committee of our University hereby grants approval to your research proposal No. IHEC/21-22/ BT-02 entitled “Evaluation of Oxidative Biomarkers in Relation with the Self-reported Symptoms among over usage of Smart Phone users on the basis of age” submitted by you. The Approval number for the same is AUW/IHEC/BT-21-22/XPD-02.</p> <p>We wish you all the best in your research endeavours.</p> <p style="text-align: right;">Regards,  Dr.S.Uma Mageshwari Member Secretary</p> <div style="text-align: right;">  </div>

Annexure 3

Paper presented on National conference sponsored by SERB and CSIR

ABSTRACT BOOK

NATIONAL CONFERENCE ON
"Recent Advances in Life Sciences towards Health Care"
(RALSH-2022)

Sponsored by

SERB & CSIR

22nd & 23rd April, 2022

Organized by

SCHOOL OF LIFE SCIENCES (Ooty Campus)



JSS ACADEMY OF HIGHER EDUCATION
& RESEARCH, MYSURU
(Deemed to be University)
Accredited 'A+' Grade by NAAC

OP-29

**A SURVEY ON EXTENSIVE USAGE OF MOBILE PHONE AND ASSOCIATED
COMPLICATIONS BASED ON AGE**Shirley A¹ and Shalini K.V²*¹Student, II M.Sc Biotechnology, Department of Biochemistry, Biotechnology and Bioinformatics,
Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore,**²Assistant Professor, Department of Biochemistry, Biotechnology and Bioinformatics,
Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore*Corresponding author: Mail ID: shirley.anthony.9212@gmail.com

Mobile phone is the most dominant portal of information and communication technology. It is used for education, entertainment, playing games, social network, online shopping, gambling, business, reading books, communication, photography, videography, etc. At the same time over usage of mobile phones may cause so many complications like eye irritation, wrist pain, eye dryness, weakness of thumb and wrist, neck pain, computer vision syndrome, insomnia, sleep disturbance, stress, hallucination, etc. Hence an attempt has been made to analyse the extensive usage of mobile phone and complications caused among the people through questionnaire. Questionnaire was prepared and online survey method was carried out to collect the data including demographic details, habituation, previous awareness on mobile phone hazards and addiction behaviour of the participants. Statistical analysis was performed using SPSS 25 to analyse the complication of over usage of mobile phone among participants based on the age and the results were summarized. From the survey, it is notable that all the age groups are using mobile phones. But children and young people are addicted too much to the mobile phone than the aged people.

Key points: Questionnaire, demographic details, statistical analysis