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Phytochemical profiling and anti-inflammatory activity of selected nutritional seeds

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ABSTRACT:

Seeds are the great sources of fibre. They contain healthy monounsaturated fats, polyunsaturated fats and many important vitamins, minerals, anti-inflammatory and antioxidants. In the present investigation, the phytochemical analysis and anti- inflammatory of *Helianthus annuus* L., (Sunflower) *Ocimum basilicum* L. (Basil) and *Oryza sativa* L. *indica* (Black rice) were carried out. Qualitative screening revealed the presence of alkaloids, glycosides, tannin and phenol, carbohydrate, saponin, anthocyanin, betacyanin, steroid, protein, coumarin, flavonoid, fat and oil. Among 16 phytochemical constituents, sunflower seed have 11 compounds when compared to basil seed and black rice. Sunflower seed are highly nutritious when compared to other two. In vitro anti-inflammatory assay of sunflower seed, basil seed and black rice extracts at different concentration were studied by protein denaturation. At 100mg concentration basil seed exhibit higher level of inhibition (48.3%) when compared to sunflower (47.9%) and black rice (46.6%). This suggests that seeds can be used as food supplements in every day diet for normal metabolic activities of living organism, to reduce the anti-nutritional effects and also useful in pharmaceutical industry.

Keywords: Nutritive seed, Phyto-chemical analysis, Anti-inflammatory activity, Food Supplement

INTRODUCTION:

Seeds are known to be super-nutritious, powerhouse of nutrients and can be consumed daily for a myriad list of health benefits. They contain all the starting materials necessary to develop into complex plants. Because they are extremely nutritious. Seeds are high in dietary fibre and have huge potential as a functional ingredient. They are known to be extremely versatile and can be incorporated any way in any dish.

Sunflowers are botanically classified as *Helianthus annuus* L. Sunflower seeds are good source of minerals. Sunflower seeds are among the best source of vegetable proteins, and their

nutritional and functional properties (Venktesh and Prakash, 1993). Sunflower seed contains an appreciable amount of oil. Sunflower oil is the non-volatile oil expressed from sunflower seeds. The oil is important with respect to its fatty acid profile and tocopherols contents. Tocopherols are natural antioxidants. A tiny sunflower seed is a package of healthy unsaturated fats, protein, fibre and other important nutrients like vitamin E, selenium, copper, zinc, folate, iron and phytochemicals.

Ocimum basilicum L., commonly known as basil or sweet basil, is an annual spicy herb of the Labiatae family. Basil seeds are black in color and with oval shape and are good source of fiber. Basil seeds is a good source of iron, calcium, and magnesium. The mucilage extracted from basil seeds has emulsifying, foaming, thickening, stabilizing, viscosity, and gelling properties (Naji-Tabasi and Razavi, 2016). Basil has also been widely used in traditional medicine in the treatment of headaches, coughs, constipation, diarrhea, warts, worms, and kidney problems (Simon *et al.,* 1999). In addition, various pharmacological actions have been described, such as stomachic, antioxidant, antiviral, antimicrobial, analgesic, anti-inflammatory, antidiabetic, and anti-stress activities, and antipyretic diuretic and emmenagogue properties (Bilal *et al.,* 2012*,* Nadeem *et al.,* 2020).

Black rice is a type of the rice species *Oryza sativa* L. *indica* which is black in color, glutinous, packed with high level of nutrients and mainly cultivated in Asia. Black rice has minerals (Ca, P, Fe, and Zn) and dietary fiber, which are higher than brown and white rice. The pericarp (outer part) of kernel of this rice color is black due to presence of high amount of a powerful pigment known as anthocyanins, which are flavonoids that perform as antioxidants in the body. Black rice is a super nutritious type of rice that is high in fiber, antioxidants, vitamins B, vitamin E, iron, thiamine, magnesium, niacin and phosphorous (Kushwaha, 2016; Zhimin Xu, 2010).

Phytochemicals are naturally occurring components in fruits, vegetables, legumes and grains. They are non-nutritive plant chemicals with defensive properties against cancer by protecting the cells from damage. Most of the phytochemicals possess the biological antioxidant capacity. These phytochemicals tend to prevent the adhesion of pathogens to the human cell wall by physically binding to it (Nandini *et al*., 2020). There are more than thousand known phytochemicals. They are acting as antioxidants, stimulating enzymes, interfering with DNA replication, destroying bacteria, as well as they seem to act to reduce the onset of diseases such as cancer and heart diseases (Krishnaswamy and Raghuramulu, 1998). Fruits and vegetables are important sources of phytochemicals and it is studied that some anti-nutritional exhibited potential for reducing the risk of certain diseases in human beings (Aletor and Adeogun, 1995). These diseases include high blood pressure, heart attack, stroke, and other cardiovascular diseases. These anti-nutrients or phytochemicals carry out their healing activities by combining with vitamins or with other nutrients (Liu, 2004).

Inflammation is usually a body response to living tissue damage and to a number of systemic malfunctions including asthma, atherosclerosis, arthritis, physical injury and infection (Viljoen *et al*., 2012). It may be caused by foreign bodies, physical injury, allergens, radiation,

stress, trauma, frostbite and alcohol. Inflammation serves to isolate and eliminate infectious agents, induce repair and give protective response. Unfortunately, inflammation can lead to pain and discomfort that can persist for a long period of time (Subramanian *et al*., 2003). Inflammation is either acute or chronic inflammation. Plants have the ability to synthesize a wide variety of phytochemical compounds as secondary metabolites. In the present study an attempt has been made to investigate the phytochemical constituents and the anti-inflammatory activity of sunflower seeds, basil seeds and black rice.

METHODOLOGY:

Fresh seeds of *Helianthus annuus* L. (sunflower), *Ocimum basilicum* L. (basil) and *Oryza sativa* L. *indica* (black rice) were collected from an organic shop. Seeds were washed and it is air-dried in shade at room temperature. Sun drying is the most acquired method to dry the plant sample because it is cost-saving. A minimum temperature of 30 ˚C or higher is required and several days will be taken to eliminate the excessive moisture content of the sample (Banu and Cathrine, 2015). After the drying process, the dried plant sample must be preserved in a dry container with low humidity conditions to prevent the absorption of moisture by the dried sample. The dried sample is ground into powdered form by using a mechanical blender or mortar and pestle. Next, the extraction process is started by placing the dried sample and solvent together in a vessel.

Preparation of crude extract:

50 ml ethanol and water (aqueous) were taken into sterilized conical flasks of 100 ml capacity then added 1 gm of each dried powdered sample. It is soaked for 72 hours. Later the extracts were filtered through Whatman No.1 filter paper. The supernatants were collected separately, labelled and used for screening of various phytochemicals.

Quantitative phytochemical analysis:

Phytochemical screening unlike pharmaceutical chemicals, phytochemicals are safe and dependable, compared to costly synthetic drugs which are invariably associated with adverse effects; therefore, it is considered as “man-friendly medicine”.

a) Test for alkaloids

$Fecl_3$ test: One drop of $Fecl_3$ solution were added to each of the test sample, formation of yellow precipitate were resulted reacting positively for alkaloids (Brindha *et al*, 1977; Trease and Evans, 1989; Sofowora, 1993; Harborne, 1998; Samatha *et al*, 2012; Archana *et al*, 2012).

b) Test for glycosides:

Concentrated sulphuric acid test: Conc.H_2SO_4 were added to test sample which resulted in appearance of reddish colour (Brindha *et al*, 1977; Trease and Evans, 1989; Sofowora, 1993; Harborne, 1998; Samatha *et al*, 2012; Archana *et al*, 2012).

c) Test for tannins and phenolic compounds:

Alkaline reagent: When sodium hydroxide solution were added to the sample solution results in the formation of yellow to red precipitate (Brindha *et al*, 1977; Trease and Evans, 1989; Sofowora, 1993; Harborne, 1998; Samatha *et al*, 2012; Archana *et al*, 2012).

d) Test for carbohydrates:

Benedict's test: Crude extract mixed with 2ml of Benedict's reagent and boiled, a reddish brown precipitate formed which indicated the presence of the carbohydrates (Sofowora, 1993; Trease and Evans, 1989; Harborne, 1998).

e) Test for saponins:

Foam test: Crude extract were mixed with 5ml of distilled water in a test tube and it was shaken vigorously. The formation of stable foam were taken as an indication for the presence of saponins (Brindha *et al*, 1977; Trease and Evans, 1989; Sofowora, 1993; Harborne, 1998; Samatha *et al*, 2012; Archana *et al*, 2012).

f) Test for anthocyanin and betacyanin:

Each sample (1mg) were boiled for 10 min in 2 ml of 1 N sodium hydroxide. Formation of bluish green colour were the sign of anthocyanin and yellow colour formation of betacyanin presence (Trease and Evans, 1989).

g) Test for steroid:

Crude extract were mixed with 2ml of chloroform and concentrated H_2SO_4 was added sidewise. A red colour produced in the lower chloroform layer indicated the presence of steroids. Another test were performed by mixing crude extract with 2ml of chloroform. Then 2ml of each of concentrated H_2SO_4 and acetic acid were poured into the mixture. The development of a greenish coloration indicated the presence of steroids (Sofowora, 1993; Trease and Evans, 1989; Harborne, 1998).

h) Test for protein:

Ninhydrin test: Crude extract when boiled with 2ml of 0.2% solution of Ninhydrin, violet colour appeared suggesting the presence of amino acids and proteins (Sofowora, 1993; Trease and Evans, 1989; Harborne, 1998).

i) Test for terpenoid:

Crude extract were dissolved in 2ml of chloroform and evaporated to dryness. To this, 2ml of concentrated H_2SO_4 was added and heated for about 2 minutes. A greyish colour indicated the presence of terpenoids (Sofowora, 1993; Trease and Evans, 1989; Harborne, 1998).

j) Test for sterols:

Libermann-Buchard test: When samples were treated with a few drops of acetic anhydride, boiled and a few drops of concentrated H_2SO_4 from the sides of the test tube were added, shows a brown ring at the junction of two layers and the upper layer turns green which shows the presence of steroids (Brindha *et al*, 1977; Trease and Evans, 1989; Sofowora, 1993; Harborne, 1998; Samatha *et al*, 2012; Archana *et al*, 2012).

k) Test for coumarins:

An aliquot of each sample (1 mg/ml) were mixed with 1 ml of 10 % sodium hydroxide. The appearance of yellow colour formation in the test tube were the proof of coumarins presence in test samples (Harborne, 1998).

l) Test for anthraquinone:

Development of red colour were considered as an indication of the presence of anthraquinone after mixing 1 mg of each sample with 2 ml of diluted 2 % hydrochloric acid (Harborne, 1998).

m) Test for flavonoids:

Alkaline reagent test: Crude extract were mixed with 2ml of 2% solution of NaOH. An intense yellow colour were formed which turned colourless on addition of a few drops of diluted acid which indicated the presence of flavonoids (Sofowora, 1993; Trease and Evans, 1989; Harborne, 1998).

n) Test for fats and oils:

Saponification test: Add a few drops of 0.5N alcoholic potassium hydroxide to various extracts with a drop of phenolphthalein separately and heat on a water bath for 1-2 hours, formation of soap or partial neutralization of alkali indicates the presence of oils and fats (Brindha *et al*, 1977; Trease and Evans, 1989; Sofowora, 1993; Harborne, 1998; Samatha *et al*, 2012; Archana *et al*, 2012).

o) Test for quinones:

Alcoholic KOH test: When alcoholic KOH were added to the test samples red to blue colour appears reacting positively for quinones (Brindha *et al*, 1977; Trease and Evans, 1989; Sofowora, 1993; Harborne, 1998; Samatha *et al*, 2012; Archana *et al*, 2012).

In vitro assay of anti- inflammatory:

In this assay bovine serum albumin (BSA) is used as a protein. Denaturation of protein is induced by keeping the reaction mixture at 70 °C in a water bath for 10 minutes (Mizushima and Kobayashi,1968). A reaction mixture consists of various concentrations of seed extract 1000µL (50,100 µg/ml). 450 µL of bovine serum albumin is added to each concentration and incubated at 37 °C for 20 minutes and 2000 µL of phosphate saline buffer were added to each mixture and

heated at 80°C for 5 minutes. After cooling under running tap water, their absorbance is measured at 770nm. The percentage inhibition is calculated using the following equation

$$\% \text{ of inhibition} = \frac{\text{Abs of control - Abs of sample}}{\text{Abs of control}} \times 100$$

RESULT AND DISCUSSION:

Phytochemical analysis:

Qualitative screening revealed the presence of alkaloids, glycosides, tannin and phenol, carbohydrate, saponin, anthocyanin, betacyanin, steroid, protein, coumarin, flavonoid and fat and oil as given in Table 1. Phytochemical analysis of aqueous and ethanol extracts of *Helianthus annuus* L. seeds revealed the presence of alkaloids, glycosides, tannin and phenol, carbohydrate, saponin, betacyanin, steroid, protein, coumarin, flavonoid and fat and oil, whereas anthocyanin, terpenoid, sterol, anthraquinone and quinone were completely absent. Tannin and phenol, coumarin and flavonoid were strongly positive in the tests performed by aqueous and ethanol extracts. Tests conducted for the presence of tannin and phenol and coumarin gave positive results in aqueous extracts while in other ethanol extract, they were weakly detected. Tests performed for the presence of alkaloids, glycosides, carbohydrate, saponin, betacyanin, steroid, protein and fat and oil were screened positive in ethanol extract whereas aqueous extract were found to be negative.

Phytochemical analysis of aqueous and ethanol extracts of seeds of *Ocimum basilicum* L. revealed the presence of alkaloids, glycosides, flavonoid, carbohydrate, anthocyanin and steroid, whereas tannin, phenol, protein, saponin, terpenoid, sterol, coumarin, anthraquinone, fat, oil and quinone were completely absent. Alkaloids, glycosides, carbohydrate, anthocyanin and flavonoid were strongly positive in the tests performed by aqueous and ethanol extracts. Tests conducted for the presence of alkaloid, carbohydrate and flavonoid gave positive results in aqueous extracts while in other ethanol extract, they were weakly detected. Tests performed for the presence of anthocyanin was screened positive in ethanol extract whereas aqueous extract were found to be negative.

Phytochemical analysis of aqueous and ethanol extracts of seeds of *Oryza sativa* L. *indica* revealed the presence of alkaloids, glycosides, flavonoid, carbohydrate, anthocyanin and steroid, whereas tannin, phenol, protein, saponin, terpenoid, sterol, coumarin, anthraquinone, fat, oil and quinone were completely absent. Alkaloids, glycosides, carbohydrate, anthocyanin and flavonoid were strongly positive in the tests performed by aqueous and ethanol extracts. Tests conducted for the presence of alkaloid, carbohydrate and flavonoid gave positive results in aqueous extracts while in other ethanol extract, they were weakly detected. Tests performed for the presence of anthocyanin were screened positive in ethanol extract whereas aqueous extract were found to be negative.

Among 16 phytochemical constituents, sunflower seed have 11 compounds when compared to basil seed and black rice. Sunflower seed are highly nutritious when compared to

other two. Phytochemicals are bioactive compounds found in parts of plants, fruits and vegetables. They contribute to the colour, aroma, and flavour and protect plants from environmental hazards such as pollution, stress, drought, ultraviolent (UV) exposure and pathogenic attack (Christensen *et al*., 2013). More importantly, it is believed that they are responsible for the protective, ameliorative and therapeutic effects associated with consumption of plant products. This is because many of them possess antioxidant, anti-inflammatory and anti-microbial activities (Aderonke *et al*., 2021).

Phytochemicals are also chemically diversified group of compounds produced by plant as a part of defence against pathogens, predators and competitors. They have specific chemical properties that are useful for humans in treatment of diseases (Halliwell, 2007). Phytochemicals responsible for broad spectrum of bioactivities include alkaloids, glycosides, tannins, polyphenols, flavonoids and many more (Joseph and Raj, 2010).

The phenolic compounds are one of the largest and most ubiquitous groups of plant metabolites (Singh *et al*., 2007). They possess biological properties such as anti-apoptosis, antiaging, anti-carcinogen, anti-inflammation, anti-atherosclerosis, cardiovascular protection and improvement of endothelial function, as well as inhibition of angiogenesis and cell proliferation activities (Han *et al*., 2007).

The study of *Helianthus annuus* L. seeds, *Ocimum bacilicum* L. seeds and *Oryza sativa* L. *indica* seeds revealed the presence of phyto-constituents, preliminary phytochemical screening of the different fractions of dried fruit extract of *Coccinia grandis* were also revealed the presence of alkaloids, carbohydrates, phytosterols, tannins, flavonoids, protiens and diterpenes. The type of phyto-constituents of methanol, ethanol, chloroform, n-hexane and petroleum ether in *Coccinia grandis* (Ashwini *et al*., 2012).

The analysis of *Artemisia* species showed the presence of coumarin, saponins, glycosides, flavanoids, proteins, and triterpenoids (Wright, 2002). There was also report that terpenoids is one of the most commonly found metabolites in most of the genus of *Artemisia* (Tanaya *et al*., 2013). A similar phytochemical study in the aerial parts of *Artemisia vulgaris* had shown the presence of certain secondary metabolites like amino acid, carbohydrate, flavonoids, phenolic compounds, phytosterol, proteins, saponins and tannin (Ashok *et al*., 2010).

Phytochemical constituents such as flavonoids, alkaloids, tannins are present in ethanol extracts. Carbohydrates is present in both the chloroform and ethanol extracts of *Murraya koenigii* L. amino acids and phytosterols were present in the chloroform extracts (Gandhi and Dhinek, 2021).

Phytochemical screening of methanol extract of *Peperomia pellucida* revealed the presence of alkaloids, flavonoids, saponin, cardiac glycosides, tannins, triterpenoids, steroids, xanthoproteins, phlobatannins but anthraquinone were found to be absent. This is in agreement with earlier observations of Egwuche *et al*., 2011 and Gini and Jothi, 2013 who also observed the presence of all the above phytoconstituents except anthraquinone from the extracts of *P. pellucida*. The same result i.e. the absence of anthraquinone in aqueous and ethanol extracts were revealed in *Helianthus annuus* L., *Ocimum basilicum* L. and *Oryza sativa* L. *indica*. The

presence or absence of a phytoconstituent as well as its quantity depend on a number of factors such as place of collection, time of collection, period of collection as well as extraction methods and solvent employed in the extraction. Moreover, studies have shown that hydroalcoholic tends to extract more active biomolecules than aqueous or other solvents.

Table:1 Phytochemical constituents of sunflower seed, basil seed and black rice

Phytochemical constituents	Tests	Sunflower seed		Basil seed		Black rice	
		Aqueous extract	Ethanol extract	Aqueous extract	Ethanol extract	Aqueous extract	Ethanol extract
Alkaloids	$FeCl_3$ test	-	+	-	+	+	+++
Glycosides	Con.H_2SO_4 test	-	+	-	+	++	-
Tannin\Phenol	Alkaline test	+++	++	+	+	-	-
Carbohydrate	Benedict's test	-	+	-	-	+++	+
Saponin	Foam test	+	-	+	+	-	-
Anthocyanin	Anthocyanin test	-	-	-	-	+	+++
Betacyanin	Betacyanin test	+	-	++	+	-	-
Steroid	Steroid test	-	+	-	-	+	-
Protein	Ninhydrin test	-	+	+++	+	-	-

Terpenoid	Terpenoid test	-	-	-	-	-	-
Sterol	Libermann - Buchard test	-	-	-	-	-	-
Coumarin	Coumarin test	+++	++	+	-	-	-
Anthraquinone	Anthraquinone test	-	-	-	-	-	-
Flavonoid	Alkaline reagent test	++	+++	-	-	+	+
Fat & Oil	Saponification test	-	+	-	-	-	-
Quinone	Alcoholic KOH test	-	-	-	-	-	-

Anti-inflammatory assay:

Anti-inflammatory potential of ethanol extracts of *Helianthus annuus* L., *Ocimum basilicum* L. and *Oryza sativa* L. *indica*. were evaluated against denaturation of egg albumin as depicted in Table 2. The present findings exhibited a concentration depended inhibition of protein denaturation.

In vitro anti-inflammatory assay of sunflower seed, basil seed and black rice extracts at different concentration were studied by protein denaturation. The percentage of inhibition exhibited by sunflower seed extract concentration in 100mg has higher level of inhibition (47.9%) whereas the seed concentration at 50 mg showed lower level of inhibition (31.8%), by basil seed extract concentration in 100mg has higher level of inhibition (48.3%) whereas the seed concentration at 50 mg showed lower level of inhibition (32.7%) and by black rice extract concentration in 100mg has higher level of inhibition (46.6%) whereas the seed concentration at 50 mg showed lower level of inhibition (30.7%). At 100mg concentration basil seed exhibit higher level of inhibition (48.3%) when compared to sunflower (47.9%) and black rice (46.6%).

Denaturation of proteins is a well-documented cause of inflammation. Protein denaturation occurs when proteins lose their biological functions due to the change in their structures. This happens due to other compounds, heat, or any other external stress. Therefore, the denaturation of tissue proteins is considered one of the markers of inflammation.

Abdallah *et al*., 2014, worked on the anti-inflammatory activity of *Trichodesma trichodesmoides* var. *tomentosum* and found the butanol fraction as more the most potent in showing the highest anti-inflammatory activities than their individual fractions. Sukumaran and Yadav 2016, worked on anti-inflammatory potential of *Dendrobium macrostachyum* Lindl. where they observed that the ethanol and water extract was highly effective as albumin

Sl.No	Concentration	Sunflower seed		Basil seed		Black rice	
		OD at 770nm	**% of inhibition**	**OD at 770nm**	**% of inhibition**	**OD at 770nm**	**% of inhibition**
1	Control	1.252	-	1.252	-	1.252	-
2	50	0.853	31.8	0.934	32.7	0.725	30.7
3	100	0.971	47.6	1.23	48.3	0.872	46.6

Table:2 Anti-inflammatory assay of sunflower seed, basil seed and black ricedenaturation inhibitors (IC50, 114.13 and 135.818 µg/ml, respectively) and proteinase inhibitors (IC50, 72.49 and 129.68 µg/ml, respectively).

The inflammatory drugs (salicylic acid, phenylbutazone etc) have shown dose dependent ability to thermally induced protein denaturation. Similar results were observed from many reports from plant extract of *Oxalis corniculata* (Sakat *et al*., 2010). The extracts may possibly inhibit the release of lysosomal content of neutrophils at the site of inflammation. These neutrophils lysosomal constituents include bactericidal enzymes and proteinases, which upon extracellular

release cause further tissue inflammation and damage (Chou 1997). Proteinases have been implicated in arthritic reactions. Neutrophils are known to be a source of proteinase which carries in their lysosomal granules many serine proteinases. It was previously reported that leukocytes proteinase play important role in the development of tissue damage during in inflammatory reactions and significant level of protection were provided by proteinase inhibitors (Das and Chatterjee, 1995). Recent studies have shown that many flavonoids and related polyphenols contributed significantly to the antioxidant and anti-inflammatory activities of many plants (Tubaro *et al*., 1988). Hence, the presence of bioactive compounds in the water extract of different parts of *Wedelia trilobata* may contribute to anti-inflammatory activity (Govindappa *et al*., 2011).

Esculetin (6,7-dihydroxy-2H-1-benzopyran- 2-one) is a coumarin derivative found in various natural plant products and has been reported to have beneficial biological and biochemical activities. For example, esculetin has been shown to have an anti-inflammatory effect in the croton oil ear test (Tubaro *et al*., 1988).

According to *Calligonum comosum* the results of protein denaturation the methanol and flavonoid extracts show the best values compared to other extracts. The difference between the three extracts was statistically highly significant in the albumin denaturation test ($p < 0.001$) (Chouikh *et al*., 2020). Protein denaturation is a process in which proteins lose their tertiary structure and secondary structure by the application of external stress such as a strong acid or base, a concentrated inorganic salt, an organic solvent, or heat (Anoop and Bindu, 2015). Denaturation of proteins is one of the well documented causes of inflammation and leads to various inflammatory diseases (Osman *et al*., 2016). The possible mechanism of denaturation is the alteration of electrostatic, hydrogen, hydrophobic, and disulphide bonds which maintain the three-dimensional structure of proteins (Chirisa and Mukanganyama, 2016). The anti-denaturing activity of our extracts may be due to the interaction of certain components with two sites (present in certain proteins such as albumin) of bonds rich in tyrosine, threonine, and lysine (Williams *et al*., 2008). Kurlbaum and Hogger, 2011 reported that certain phenolic compounds such as flavonoids and phenolic acids possess the ability to bind to plasma proteins.

Flavonoid-rich compounds have been shown to exhibit very high anti-inflammatory potentials. The mode of response of the erythrocyte was monophasic, in agreement with the earlier observations of Oyedapo *et al*., 1999 in the investigations of red blood cell membrane stabilizing potentials of extracts of *Lantana camara* and its fractions. The assay of albumin denaturation showed that there was inhibition of albumin denaturation at various concentration of the *Peperomia pellucida* methanol extract and standard drug. The standard drug (diclofenac) showed the highest percentage inhibition when compared with *Peperomia pellucida* methanol extract (Aderonke *et al*.,2021). This also was in line with the observations of Aina and Oyedapo 2013, who also reported the inhibition of albumin at various concentrations of the extracts and standard drug. It is therefore possible that the anti-inflammatory potential of *Peperomia pellucida* methanol extract might be attributed to the phytochemicals present in it.

The anti-inflammatory activity of *Murraya koenigii* at different concentrations increases with increasing concentration (Gandhi and Dhinek, 2021). The present study of *Helianthus annuus* L. seed also results that the increase with increasing concentration. Among the plants of Balochistan, *Ferula oopoda* the Apiaceae family has a great representation, and several species are used because they contain antimicrobial compounds, anti-inflammatory and antifungal agents (Chanda and Rakholiya, 2011). Anthraquinones, coumarins, and anthocyanin are endowed with many biological activities like antimicrobial, antioxidant, anti-inflammatory and anticancer (Chen *et al*., 2007; Garazd *et al*., 2007; Ifesan *et al*., 2009; He *et al*., 2011; Amiery *et al*., 2012; Choi *et al*., 2013).

Lophira procera A. Chev results showed inhibition of protein denaturation (albumin) dependent on the concentration of aqueous extract. The aqueous extract were used with concentrations ranging from 31.25 to 500 lg/mL, the percentage inhibition increased with the concentration ranging from 230 ± 0.531% to 4360 ± 1.078%. Diclofenac sodium, used at concentrations ranging from 156.25 to 2500 lg/mL, were used as a reference drug which also showed inhibition of concentration-dependent protein denaturation. The effect of diclofenac

sodium were found to be rather low compared to that of the aqueous extract tested (Leonid *et al*.2017).

CONCLUSION:

Health and nutrition rank high for consumers who wants food that are as good for them as they are good to eat. Sunflower seeds, basil seeds and black rice meet the challenge with their combination of nutrients and health benefits. The present screening of phytochemical in *Helianthus annuus* L. (sunflower seed), *Ocimum basilicum* L. (Basil seed) and *Oryza sativa* L. *indica* (Black rice) posses alkaloids, glycosides, tannin and phenol, carbohydrate, saponin, anthocyanin, betacyanin, steroid, protein, coumarin, flavonoid and fat and oil. This suggests that seeds can be used as food supplements in every day diet for normal metabolic activities of living organism to reduce the anti-nutritional effects and where are also useful in pharmaceutical industry. According to the phytochemicals and bioactivities studied, the seeds of *Helianthus annuus* L. *Ocimum basilicum* L. and *Oryza sativa L. indica* has the greatest anti- inflammatory potential with the highest alkaloid and protein content. More so, the seeds were found to exhibit a wide range of anti-inflammatory potential enabling it to be useful in the management of several disease condition. Hence, more work is possible on the above plants to reveal the unknown important which would be helpful, which is the need of an hour for the present pharmaceutical world.

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