



Characterization and Evaluation of Anti-Diabetic, Anti-Inflammatory Activity, in Experimental Animals and Screening of *Brassica rapa* Components

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Abstract: Volatiles are good leading pointers towards their antecedents and hence the quality of the item can be established. Low levels of aroma volatile synthesis in immature stages of the plant are suggestive of low precursor levels prevailing which are gradually controlled until the correct harvest time. A substantial number of the primary and secondary metabolites extracted from the leaves of *Brassica rapa* are fundamental in the improved human nutrition. Powder of *Brassica rapa* leaves was prepared from shade dried and pulverised leaves of different plantlets. One hundred grams of the powder was placed in soxhlet device ready for continuous hot percolation for approximately 8 hours using methanol with a volume of 350ml as a solvent. This was done under vacuum first to a semi-solid paste and then, dried further in desiccator to remove the residual solvent contents. The main phytochemicals identified in leaves were 3-tert-Butylphenylisothiocyanate, $C_{11}H_{13}NS$, Sinapinic acid-O-glucuronide isomer, $C_{17}H_{20}O_{11}$, Pentenyl-1,3-dioxan-5-ol, $C_9H_{16}O_3$, 9-Octadecenamide, $C_{18}H_{35}NO$, Cyanidin 3-O-(6-O-malonyl-beta-D-glucoside, $C_{24}H_{23}O_{14}$, 5-Methylhex-5-enenitrile, $C_7H_{11}N$, 1-Phenylethyl isothiocyanate, C_9H_9NS , 4-methylsulfinylbutyl glucosinolate, $C_{12}H_{23}NO_{10}S_3$, 5-Methylhex-5-enenitrile, $C_7H_{11}N$, Cyanidin 3-O-(6-O-malonyl-beta-D-glucoside, $C_{24}H_{23}O_{14}$, 1-Phenylethyl isothiocyanate, C_9H_9NS , n-Hexadecane, $C_{16}H_{34}$, Glucoraphanin, $C_{12}H_{23}NO_{10}S_3$, Phytol, $C_{20}H_{40}O$, and 3,3'-Diindolylmethane, $C_{17}H_{14}N_2$. Impact of oral administration of *Brassica rapa* fractions on serum enzymes in rats: To ascertain the effect of oral administration of *Brassica rapa* extract on the serum enzymes SGPT, SGOT, and ALP, laboratory rats were used in in vitro experimental testing. Recorded 79.15 ± 1.66 , 95.17 ± 2.90 and 35.92 ± 0.87 respectively for *Brassica rapa* methanol extract, while 139.00 ± 5.50 , 154.81 ± 6.20 and 41.93 ± 1.05 were recorded respectively for using Di-(2-ethylhexyl) phthalate and 52.97 ± 1.20 , 70.00 ± 1.62 and 19.00 ± 0.03 were recorded to Control (vehicle) (0.5 ml/kg Corn oil). According to the type of extract (Methanolic crude extract, Hexane fraction, Ethanol fraction, Water fraction and acarbose (Standard) recorded 93.02 ± 0.65 , 38.01 ± 0.37 , 55.91 ± 0.29 , 69.88 ± 0.47 and 14.11 ± 0.05 respectively inhibitory potency against α -amylase. While recorded 67.44 ± 0.43 , 46.07 ± 0.30 , 35.02 ± 0.27 , 27.00 ± 0.21 , and 11.96 ± 0.06 respectively inhibitory potency against α -glucosidase activity. *Brassica* extract's diverse patterns of bioactive chemicals make it a potential natural anti-diabetic and source for a number of other medical uses. We assessed *Brassica rapa* leaves' anti-inflammatory, anti-diabetic, and nutritional qualities. The study further reveals that this plant's leaves contain ascorbic acid, phenolic compounds, carotenoids and flavonoids which are all beneficial to health. It should be noted that this plant's leaves provide unique phenolic compounds. When biological activities are taken into account, this could be quite relevant and merits more research. These plant components' unique chemical makeup greatly influences their anti-inflammatory qualities. Therefore, the food and pharmaceutical companies may consider the commercialisation of standardised aqueous extracts of leaves for use as antidiabetic.

Keywords: Anti-Diabetic, Anti-Inflammatory, Experimental animals, *Brassica rapa*

Introduction

Brassica rapa is renowned for its seed oil and green vegetables. This plant is used to treat numerous chronic and non-communicable diseases because of its nutraceutical potential, which includes anti-inflammatory, anti-carcinogenic, antiulcer, and anti-cardiovascular qualities. Because of its high nutritional content, it is a widely consumed crop with edible and beneficial portions (flashy leaves, seeds, and flowers) all over the world. Furthermore, it is a valuable crop for the production of industrial and edible oil [1–3]. Numerous phytochemicals, including phenolic compounds, secondary metabolites, vitamins, antioxidants, and more, are abundant in it. The phytochemicals included in this crop also provide strong, all-around protection against the common substances that cause cancer. A small number of phytochemicals are plant volatiles that primarily reduce herbivore attacks, draw pollination from animals, and may even be beneficial to human health. High-value agricultural products, seed oils can be used as industrial or fuel oils or as refined culinary oil products. The fatty acid profile of seed oils has a major role in determining their appropriateness for both human consumption and combustion. Higher proportions of unsaturated fatty acids (16–18 carbons), especially monounsaturated fatty acids (MUFAs), in seed oils make them appropriate for use as edible oil and as feedstock for the generation of biodiesel [4]. Volatile molecules have already been shown to support the organoleptic qualities of plant foods, which benefits human health. Research on identifying volatile chemicals in various crucifers has been conducted. The phenolic contents and the antioxidant potentials of different *Cyamopsis tetragonoloba* (L.) varieties have been recently elucidated through employing high performance liquid chromatographic methods. Therefore, the current study employed GC–MS analysis of the volatile chemicals from leaves following further attempts to chemically characterize several cluster bean varieties. Among the many volatile substances produced by brassica plants are glucosinolates [5-7], terpenes, alcohols, esters, aldehydes, and ketones. These volatiles' primary function is probably to lessen the herbivorous onslaught. They may also play a role in the pollination process by attracting animals. The biological potential of plant volatiles to support human health makes them significant as well. It is actually true that the volatiles that provide flavour and affect the organoleptic qualities of plant foods are linked to health [9].

The production of various volatile chemicals via a number of additional pathways also adds to the volatile profile of plants. The lipoxygenase system and β -oxidation are the two main routes that catabolise fatty acids, which act as volatile precursors. Polyunsaturated fatty acid hydroperoxidation is catalysed by lipoxygenase (LOX). Hydroperoxide lyase (HPL) mediates one of the processes that further metabolize LOX products, converting them into volatile esters, alcohols, and aldehydes. Ketones in contrast are synthesized when fatty acid under-go β -oxidation and/or biosynthesis. It has come to our attention that terpenes in plants derive from either the 2-C-methyl-d-erythritol-4-phosphate or mevalonate pathways. Besides applications in the field of perfume production, norisoprenoids arising from the oxidative cleavage of carotenoids are essential to biological processes [10–13]. Certain previous investigations on volatiles employed the seedlings or leaves of *Brassica* species. Studying the fluctuating pattern during plant development will then enable us further comprehend the importance of these compounds in the maturation process. It also may identify the existence of other flammable matters which are beneficial to human and test their anti-inflammatory and anti-diabetic properties.

Materials and Methods

Collection, Preparation of Plant Material and Methanolic Extract

The local medical herb vendor who brought the leaves had identified them as those of the *Brassicarapa* plant. The leaves of the plant *Brassicarapa* were washed properly that there will not be any dirt on the sample and thereafter done to powder, but not compromised or washed. The powdered crude medication was sieved through a screen to gain a uniform particle size. Before solubility test the drug was dissolved in different solutions to discover its maximum solubility [38]. On the same basis but independently, leaves of the plant species *Brassicarapa* L. were dried under shade condition and made into powder form. Methanol (350 ml) was used as a solvent while 100 g of powder was

packed in a soxhlet apparatus and reflux for about 8 hours. When the extract had turned into a thick slush by vacuum concentration, it was spread on petri dishes and left to dry in a desiccator.

Gas Chromatography-Mass Spectrometry Analysis

The chemical hydrolytic products were analyzed using capillary column namely DB-5MS aneeding 30 m x 0.25 mm i. d,0.25 m and Shimadzu's (QP-2010) gas chromatography mass spectrometer in AOC-20i auto-sampler. .pyridine extraction was carried out in a GC grade DCM, and the sample injection volume was 2 l, while the column temperature programmed was 230°C at a rate of 4°C/min. The temperature was kept at this level for another fifteen-minutes period. Helium served as the carrier gas (1:50) at a sample injection splitter mode flow rate of 1.1 ml/min.

α -amylase inhibitory assay

Several minor deviations were incorporated to the standard procedure to determine the α -amylase inhibitory activities of the extract and the fractions. Twenty millilitres of α -amylase having two international units per millilitre was followed by 200 millilitres of extract and fractions containing varying concentrations of 0.5 milligramme per millilitre, and finally 500 millilograms of 6.8 phosphate buffer containing phosphate concentration of one hundred millimolar. The mixture was then aliquoted into a 96-well plate and was incubated for 20 minutes at 37 °C. During the preincubation process, the temperature set was 37°C for incubation. The above mixture was then transferred back into another incubator at 37 degrees Celsius for 30 minutes Further, 20 litres of 1% soluble starch in 100 mM phosphate buffer pH 6.8 was added as substrate. The liquid was then boiled at a constant pressure for 10 minutes after 100 litres of DNS colour reagent had been stirred in. A Multiplate Reader (MultiskaThermo Scientific, version 1.00.40) was then imposed to get an absorbance reading at 540 nanometres. The measurement was made in order to quantify the absorbance of the final mixture. Standard concentrations of acarbose varying from 0.1 to 0.5 mg/ml were employed as the control levels. To give a comparison, a material which had blossomed through no experimental processes (extracts and fractions) was synthesized simultaneously. In particular, there were three repetitions of each experiment made. The results, expressed by the applied formula, were given in the percentage of inhibition. The IC50 values were determined from graphic analysis of the enzyme activity inhibition by the varying amounts of the fractions.

The percentage of inhibition could be determined by applying the following formula:

$$\% \text{ Inhibition} = \frac{(\text{Abs}_{\text{control}} - \text{Abs}_{\text{extract}})}{\text{Abs}_{\text{control}}} \times 100$$

α -Glucosidase Inhibitory Assay

The inhibitory effect of the extract and fractions on α -glucosidase was determined by analysis. Otherwise, using several minor modifications, the traditional procedure was followed to perform the analysis. Serum samples were prepared by was pre-cooled in a 96 well plate for 15 minutes at 37 degrees centigrade. The reaction mixture also contained twenty litres of the nine different extracts and first nine fractions at a concentration of 0.500 mg/mL, ten litres of the purified alpha-glucosidase at one unit/mL, and fifty litres of the 6.8 phosphate buffer solution, at a concentration of 100 mM. Pre incubation was administrated at 37 degree centigrade. The mixture was then incubated for a further twenty minutes at thirty-seven degrees centigrade After twenty liters of P-NPG, containing a five millimolar concentration were added as a substrate. This reaction was stopped after the addition of 50 litres of a 0.1 M sodium carbonate solution. This test actually estimated the absorption of the recently released nitrophenol by using a multiplate reader; the measurement was taken at 405 nm. Meantime, acarbose was used as a standard measurement and it was detected in the sample under examination in the concentration 0.5 mg/mL. These three tests were done approximately three times for purposes of comparing the read findings. Furthermore, a control experiment was conducted simultaneously in which the chemical under investigation was not used. The given studies were carried out

three times to ensure the assessments obtained are as precise as possible. The following expression was used to quantify the α -glucosidase inhibitory activity in terms of the percentage of inhibition:

$$\% \text{ Inhibition} = (\text{Abs}_{\text{control}} - \text{Abs}_{\text{extract}}) / \text{Abs}_{\text{control}} \times 100$$

Where the symbols A control and A extract represent the absorbance of the control and fractions, respectively. The IC₅₀ values which were computed from the graphic representations represented the amounts of fractions required to inhibit enzyme activity to 50%.

Experimental Animals

The trials were performed on healthy male Wister Albino rats which were in the weight range of 175 – 225g and were obtained from the Iraqi Atomic Energy Commission. These were placed in polypropylene cages, fed on normal lab diet and tap water was made available ad lib. The temperature level in the room ranged between 23°C \pm 2°C. Before the studies, the rats were habituated to the laboratory for at least 8hrs [14].

Drug administration and animals

Sarin was tested in albino rats weighing between 175 and 225 grammes. The animals were sourced from the animal breeding house. They were housed in cages with proper ventilation, receiving normal rat diet and they were maintained under standard room conditions (23 \pm 3°C, 55-70% relative humidity and 12/12 light/dark cycle). In total, there were twelve rats, with three subgroups of four rats each. Sham receiving normal saline served as the standard control animals while maize oil which was used as vehicle control in all experiments with methanol fraction of Brassiarapa, while 100mg/kg Di-(2-ethylhexyl) phthalate was used as the positive control animal in Group I. Groups 2 and 3 received fractions of Brassica rapa at a rate of 0.50mL/kg and 0.75 mL/kg, respectively while Group 1 received Di-(2-ethylhexyl) phthalate at a dose of 100mg/kg.

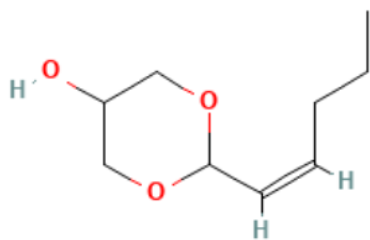
Statistical analysis

The statistical analysis was performed by using GraphPad Prism 5 Statistical Package of GraphPad Prism 5, USA. Data analysis was followed by one-way analysis of variance (ANOVA) and the Bonferroni test. For triplicate determination, the in vitro study of IC₅₀ values was expressed as mean \pm standard error mean. Phytochemicals were determined in means \pm standard deviation and free radical scavenging activities were given in percentage. As indicated by results with P < 0.05, statistical evaluation of indicator differences was taken into consideration.

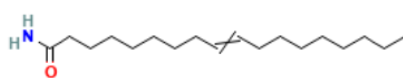
RESULTS and DISCUSSION:

An important member of the brassicaceae family of Mediterranean origin, *Brassica rapa* has been used in traditional pharmacopoeia to treat a number of chronic diseases worldwide from the earliest times. Recent literature suggests that *Brassica rapa* may be used to manage obesity and diabetes mellitus due to its ability to reduce hyperlipidemia and hyperglycemia health disorders. The primary ingredients were: 3-tert-Butylphenylisothiocyanate, C₁₁H₁₃NS, Sinapinic acid-O-glucuronide isomer, C₁₇H₂₀O₁₁, Pentenyl-1,3-dioxan-5-ol, C₉H₁₆O₃, 9-Octadecenamide, C₁₈H₃₅NO, Cyanidin 3-O-(6-O-malonyl-beta-D-glucoside, C₂₄H₂₃O₁₄, 5-Methylhex-5-enenitrile, C₇H₁₁N, 1-Phenylethyl isothiocyanate, C₉H₉NS, 4-methylsulfinylbutyl glucosinolate, C₁₂H₂₃NO₁₀S₃, 5-Methylhex-5-enenitrile, C₇H₁₁N, Cyanidin 3-O-(6-O-malonyl-beta-D-glucoside, C₂₄H₂₃O₁₄+, 1-Phenylethyl isothiocyanate, C₉H₉NS, n-Hexadecane, C₁₆H₃₄, Glucoraphanin, C₁₂H₂₃NO₁₀S₃, Phytol, C₂₀H₄₀O, and 3,3'-Diindolylmethane, C₁₇H₁₄N₂. These products are among the most unique byproducts of Brassica plants. They are used to shield them against diseases and herbivores' attacks. The extent to which glucosinolates and volatiles are produced and distributed varies depending on plant species, cultivar, vegetable part, and developmental stage of Brassica plants. On hydrolysis by its enzyme, myrosinases, glucosinolates form isothiocyanates, thiocyanates, nitriles, epithionitriles, and oxazolidines when in contact with water (in processing, cutting, tissue chewing, or wounding). A good many essential biochemical activities are stated to be

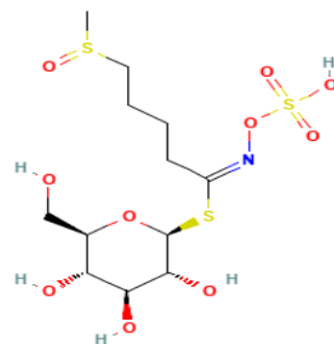
exhibited by glucosinolates and/or their products. Some are toxic when ingested by humans and animals, possibly goitrogenic, while others have beneficial effects including chemoprevention against certain types of cancer in humans.



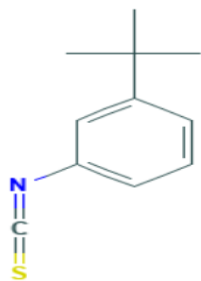
Pentenyl-1,3-dioxan-5-ol, $C_9H_{16}O_3$, 172.22 g/mol



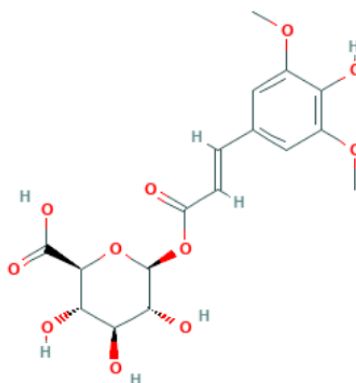
9-Octadecenamide,
 $C_{18}H_{35}NO$, 281.5 g/mol



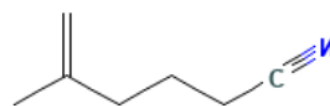
4-methylsulfinylbutyl
glucosinolate,
 $C_{12}H_{23}NO_{10}S_3$, 437.5 g/mol



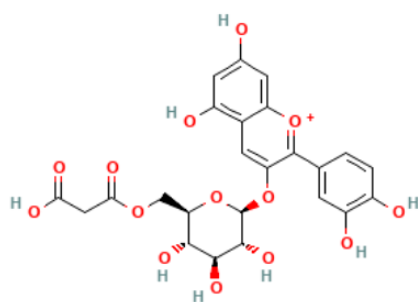
3-tert-Butylphenylisothiocyanate,
 $C_{11}H_{13}NS$, 191.29 g/mol



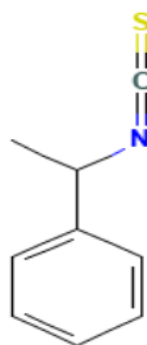
Sinapinic acid-O-glucuronide
isomer, $C_{17}H_{20}O_{11}$, 400.3 g/mol



5-Methylhex-5-enenitrile,
 $C_7H_{11}N$, 109.17 g/mol



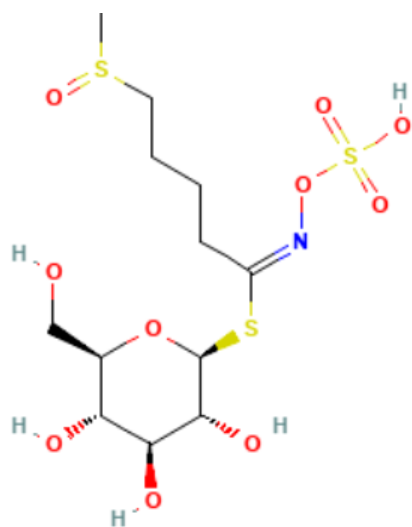
Cyanidin 3-O-(6-O-malonyl-beta-D-
glucoside, $C_{24}H_{23}O_{14+}$, 535.4 g/mol



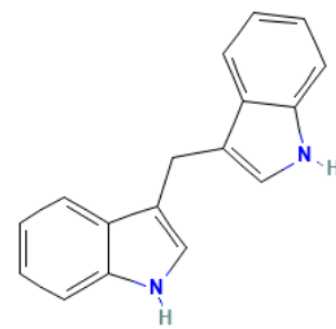
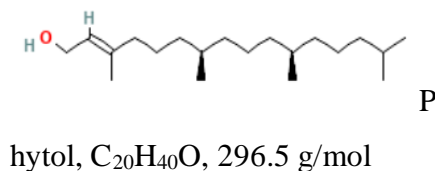
1-Phenylethyl isothiocyanate,
 C_9H_9NS , 163.24 g/mol



n-Hexadecane, $C_{16}H_{34}$,
226.44 g/mol



Glucorap



3,3'-Diindolylmethane,
 $C_{17}H_{14}N_2$, 246.31 g/mol

hanin, $C_{12}H_{23}NO_{10}S_3$, 437.5 g/mol

Impact of oral administration of *Brassica rapa* fractions on serum enzymes in rats: To find out the impact of orally administered BR extract on the rats' serum enzymes SGPT, SGOT and ALP in vivo experimental tests were conducted using laboratory rats. Recorded 79.15 ± 1.66 , 95.17 ± 2.90 and 35.92 ± 0.87 respectively for *Brassica rapa* methanol extract, while 139.00 ± 5.50 , 154.81 ± 6.20 and 41.93 ± 1.05 were recorded respectively for using Di-(2-ethylhexyl) phthalate and 52.97 ± 1.20 , 70.00 ± 1.62 and 19.00 ± 0.03 were recorded to Control (vehicle) (0.5 ml/kg Corn oil) Figure 1, 2, 3. *Brassica rapa* fractions' inhibitory efficacy against α -amylase and α -glucosidase activity: These figures show that the *Brassica rapa* had considerable anti- α -amylase and anti- α -glucosidase activities as presented in Figures 4 and 5, respectively. It was observed that the enzymatic inhibitor assay of the *Brassica rapa* fractions indicated that the inhibition activities were affected by dose as well as the fraction. The inhibition was the strongest for the highest dose under investigation, and the lowest dose gave the least inhibition. According to the type of extract (Crude extract, Hexane fraction, Ethanol fraction, Water fraction and acarbose (Standard) recorded (93.02 ± 0.65 , 38.01 ± 0.37 , 55.91 ± 0.29 , 69.88 ± 0.47 and 14.11 ± 0.05) respectively inhibitory potency against α -amylase. While recorded (67.44 ± 0.43 , 46.07 ± 0.30 , 35.02 ± 0.27 , 27.00 ± 0.21 , and 11.96 ± 0.06) respectively inhibitory potency against α -glucosidase activity. Methanol and water fraction recorded higher percentage inhibition against α -amylase than the antidiabetic drug acarbose (72.16%) at a significantly ($P < 0.05$) lower concentration. The inhibitory effects shown by methanol and ethanol fraction were significantly ($P < 0.05$) more potent than that observed with acarbose in percent inhibition of α -glucosidase. The chronic inflammatory diseases are on the rise and becoming the major causes of disability and mortality globally. The immune system which's main responsibility is to maintain the body's steady or balanced condition, of course, regulates inflammation. Since inflammation is designed to protect the body against pathogens, it is also involved in tissue repair because other endogenous stimuli including factors produced by tissue injury exist. This process involves many molecules involving mediators, for example, proinflammatory cytokines generated by macrophage including interleukin 6 (IL-6), interleukin 1 beta (IL-1 β) and tumour necrosis factor alpha (TNF- α). Acute inflammation may at times progress to chronic inflammation if the inflammatory response is not well managed. This could lead to various related diseases such as rheumatoid arthritis, type 2 diabetes, cardiovascular and neurodegenerative diseases, asthma among others [17–21]. Furthermore it has been proved that there exists a considerable relationship between inflammation and cancer and thus such aspects of pathogeny should be investigated in clinical and biomedical contexts in people. Non-steroidal anti-inflammatory drugs (NSAIDs) have been the first-line treatment among the currently available agents for inflammatory diseases because the pharmacological inhibition of cyclooxygenases that are involved in the synthesis of potent inflammatory agents such as prostaglandins [22, 23]. However, since: it is well understood that NSAIDs are associated with some toxic effects that include mainly gastrointestinal, cardiovascular, and renal toxicity the discovery of substitute drugs remains paramount. Therefore, the main subject of investigation in this case is the search for novel therapeutic agents that may be used for the prevention as well as the management of the inflammatory related disorders. Despite dedicating a lot of time to these studies, the

research community is still in need of better therapeutic agents for human inflammatory diseases that exhibit few, if any, side effects. This is because only a few of the available potentially active molecules have been taken through pharmacological or clinical application thus far. Remarkably, it has been supposed that readily available in vegetable and derivatives of it natural chemical entities are believed to contain good deal of active principles, which offer excellent prospects in the treatment or even elimination of various diseases. Besides being rich in nutrients, vegetables also contain several phytochemicals that may act as medicines [24, 25]. Hemodynamic changes, particularly endothelial disability caused by oxidative stress, can be treated by aqueous broccoli extract. Endothelial dysfunction, the first stage of cardiovascular disease, and the regulation of vascular tone may be impaired due to decreased bioavailability of NO (nitric oxide) in this syndrome. As a result, vascular disease precipitated by diabetes, hypertension or atherosclerosis could be managed with a plant extract with antioxidant capability developed in this study.

Specific biomolecules are most diverse in the members of Brassicaceae family plants and are parts of crops such as mustard greens, radishes, broccoli, and cabbages. In fact, Brassica family encompass nearly all these eight groups of phytochemicals and the GSLs. The exact manner the two interact has yet to be identified, although it has been evidenced that the DIM interferes with several signal transduction pathways that are associated with inflammatory processes and include the NFκB pathway, the PI3K pathway, PKB (often referred to as AKT kinase), and the EGFR/ERK pathway. Another similar natural compounds of concern are phenolic compounds which are commonly existing natural products that occur plant derived meals and other derived products that possess [27–31] beneficial health qualities. Such substances are enormous and complicated tannins together with derived polyphenols and simple and small molecular weight compounds and one benzene ring. The flavonoids especially the flavonols together with anthocyanins and hydroxycinnamic acid especially the chlorogenic and sinapic derivatives are the most dominant and diverse phenolic compounds in cruciferous vegetables belonging to the family Brassicaceae.

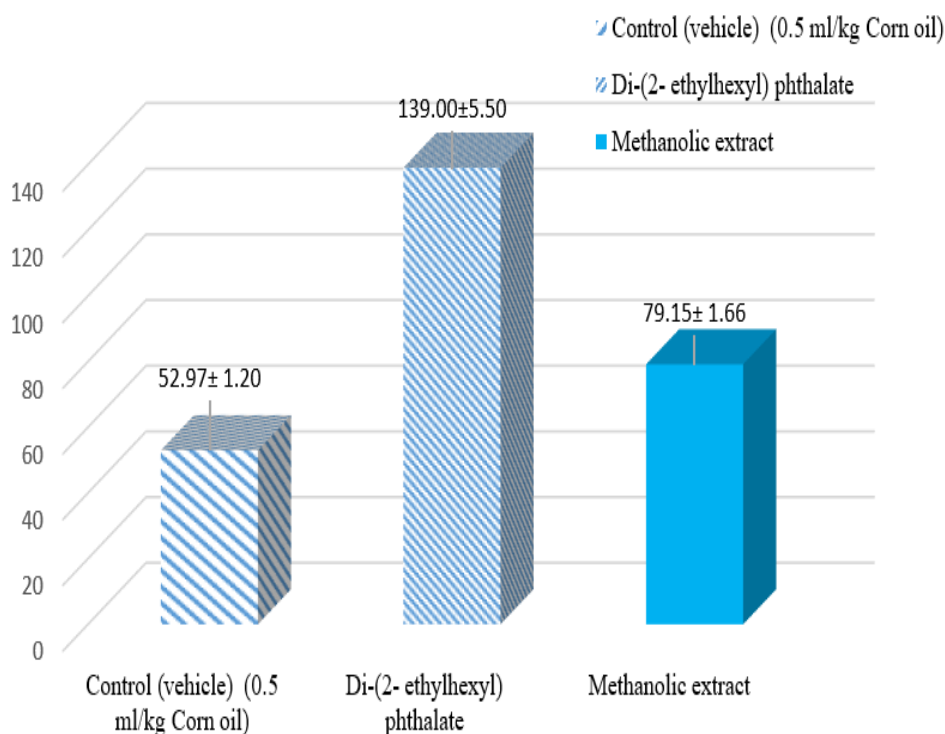


Figure 1. Oral administration of *Brassica rapa* methanolic extract, Di-(2-ethylhexyl) phthalate and Control (vehicle) (0.5 ml/kg Corn oil) on Serum Glutamate Pyruvate Transaminase

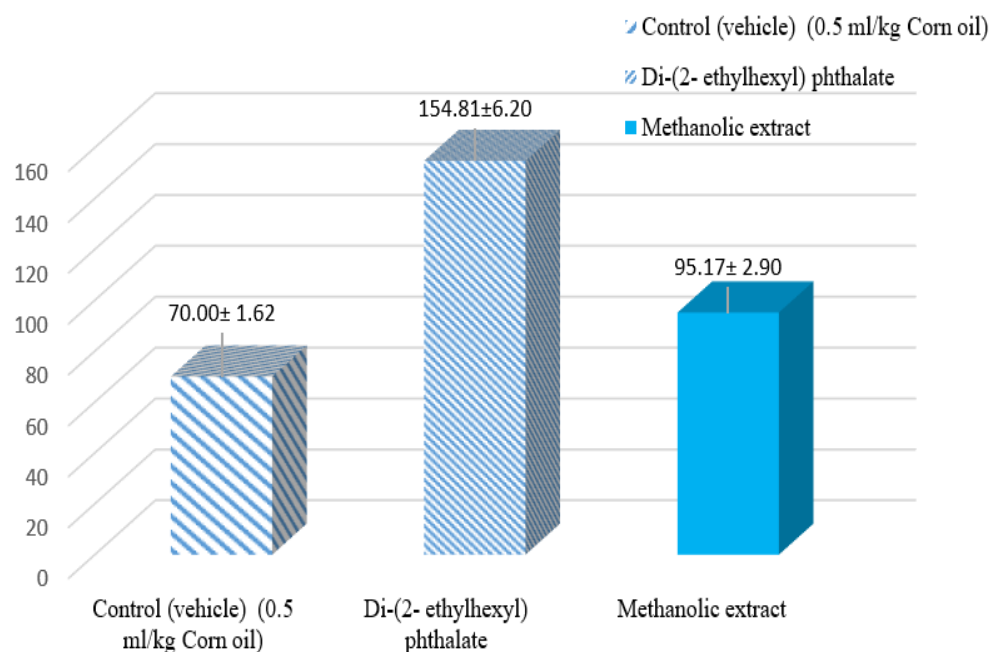


Figure 2. Oral administration of *Brassica rapa* methanolic extract, Di-(2-ethylhexyl) phthalate and Control (vehicle) (0.5 ml/kg Corn oil) on Serum Glutamic-Oxaloacetic Transaminase

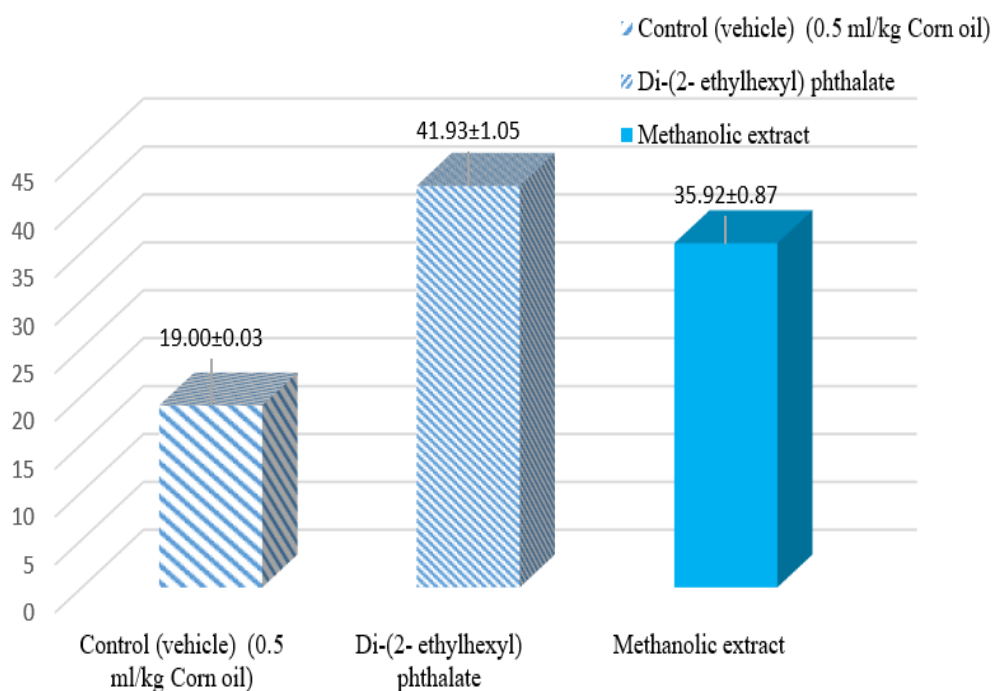


Figure 3. Oral administration of *Brassica rapa* methanolic extract, Di-(2-ethylhexyl) phthalate and Control (vehicle) (0.5 ml/kg Corn oil) on Serum Alkaline phosphatases (ALPs)

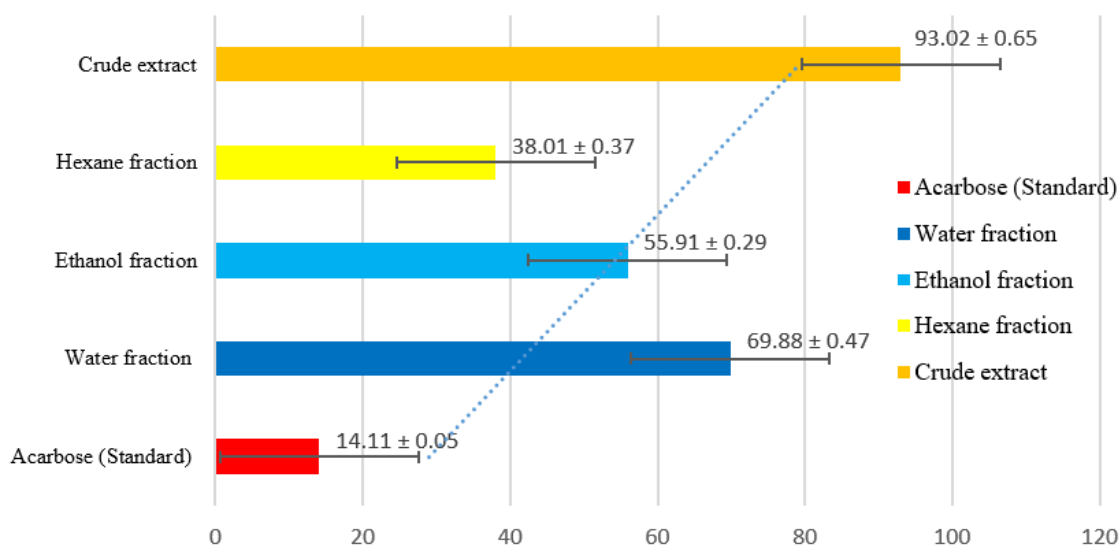


Figure 4. Inhibitory potency of *Brassica rapa* Crude, Ethanol fraction, Hexane fraction, Water fraction and acarbose as standard against α -amylase activity

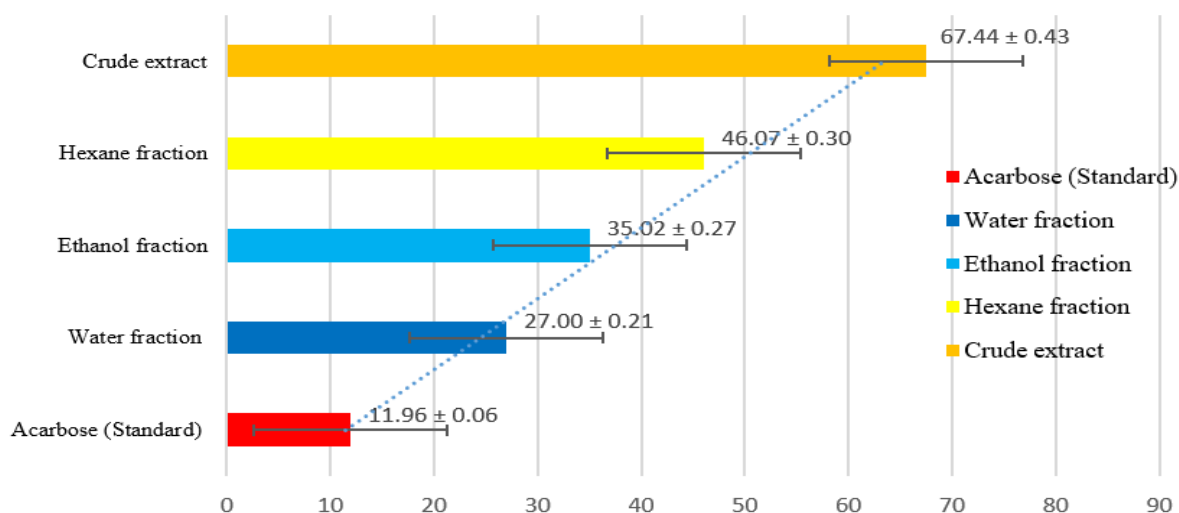


Figure 5. Inhibitory potency of *Brassica rapa* Crude, Ethanol fraction, Hexane fraction, Water fraction and acarbose as standard against α -glucosidase activity

Perhaps *Brassica rapa* possesses antidiabetic effects by inhibiting intestinal amylase enzyme, stimulating the pancreatic B cell regeneration or enhancing insulin amount. Two of the possible ways by which the extract modulates its antidiabetic effects include; easing up the distribution of blood glucose through the improved secretion of insulin by the remaining islets of Langerhans present in the pancreases. Our results revealed that the ethanolic extract of *Brassica rapa* possesses hypoglycemic effect comprising of an increasingly sharp decline in insulin and blood sugar levels [32–35]. Since the extract consists of insulin-tropic compounds, the residual beta cells are spared from destructing breakdown and new beta cells map grows. In people acute inflammation is sudden, quickly worsening, and typically lasts for a few days. If the inflammation occurs for a shorter time than chronic inflammation but a longer time than acute inflammation, it is termed subacute inflammation It may range from 2-6 weeks. Hence, any inflammation lasting a period of over six weeks is considered chronic. Low-grade inflammation that lasts over short durations of hours to days are also associated with chronic inflammation [36, 37]. Considering that mice are living comparably to humans in average, one would expect that chronic inflammation would hit rodents way faster than by

mere 6 weeks. The authors do not, however, point at which time the animal's inflammation becomes chronic. While the first can span weeks, months, or years, acute should occur only for a few days or weeks at most, based on the best and most accurate data they have.

CONCLUSION

Employing GC-MS in this study, over eleven chemical compounds were identified from the polar extract of *Brassica rapa*. Phytosterols as well as other bioactive compounds are the main property that contributes to better understanding of *Brassica rapa* uses in medicine and pharmacy. Based on the identified phytochemicals as well as their postulated biological functions, this crop may well be of great use in ethnomedicine. The results we have obtained in the present study show the potent anti-inflammatory and antidiabetic properties of different extracts of *Brassica rapa*. Furthermore, this study also provides evidence that water and methanol extract of *Brassica rapa* in rat models has dose dependant hypotensive property for the first time which provides logical justification for the use of this medicinal plant in the medical treatment. From these findings, more research is conducted on the further characterization of the molecular properties of the extracts further research on the biological profile of the extracts, where the identification and isolation of the main constituents of each extract, and an exposition of how each of the extracts operates. Essentially, more efficacy and safety investigation must be conducted on *Brassica rapa* and its major functional ingredients for use in various pathological disorders.

REFERENCES

1. Park SK, Ha JS, Kim JM, Kang JY, Lee du S, Guo TJ, et al. Antiamnesic Effect of Broccoli (*Brassica oleracea* var. *italica*) Leaves on Amyloid Beta (A β)1-42-Induced Learning and Memory Impairment. *J Agric Food Chem*. 2016;64(17):3353-61
2. Vinha AF, Alves RC, Barreira SV, Costa AS, Oliveira MB. Impact of boiling on phytochemicals and antioxidant activity of green vegetables consumed in the Mediterranean diet. *Food Funct*. 2015;6(4):1157-63.
3. Yang Y, Zhang X. In vitro antitumor activity of broccolini seeds extracts. *Scanning*. 2011;33(6):402-4.
4. Vinodh K, Natarajan A, Devi K, Senthilkumar B. Chemical composition of aqueous leaf extract of *Murraya Koenigii*. *Int J Pharm Biol Archiv*. 2013;4:493-7.
5. Gani A. Chemical constituents and uses, medicinal plants of Bangladesh. *Asiat Soc Bangladesh*. 2003;434
6. Fernie AR, Trethewey RN, Krotzky AJ, Willmitzer L. Metabolite profiling: from diagnostics to systems biology. *Nat Rev Mol Cell Biol*. 2004;5(9):763-9.
7. Kinnula VL, Crapo JD. Superoxide dismutases in malignant cells and human tumors. *Free Radic Biol Med*. 2004;36(6):718-44.
8. Sas K, Robotka H, Toldi J, Vecsei L. Mitochondria metabolic disturbances, oxidative stress and the kynurenin system, with focus on neurodegenerative disorders. *J Neurol Sci*. 2007;257(1-2):221-39.
9. Singh U, Jialal I. Oxidative stress an atherosclerosis. *Pathophysiology*. 2006;13(3):129-42.
10. Arteel GE. Oxidants and antioxidants in alcohol- induced liver disease. *Gastroenterology*. 2003;124(3):778-90.
11. Bolton JL, Trush MA, Penning TM, Dryhurst G Monks TJ. Role of quinones in toxicology. *Chem Res Toxicol*. 2000;13(3):135-60.
12. Guidi I, Galimberti D, Lonati S, Novembrino C Bamonti F, Tiriticco M, et al. Oxidative imbalance in patients with mild cognitive impairment and Alzheimer's disease. *Neurobiol Aging*. 2006;27(2):262-9.
13. Hyun DH, Hernandez JO, Mattson MP, de Cabo R. The plasma membrane redox system in aging. *Ageing Res Rev*. 2006;5(2):209-20.
14. Al-Baoqai, N. Al- mahbashi, H.; Al-Adhal, A. Antidiabetic and an-[19] tihyperlipidemic activity of dracaena cinnabaribalf resin ethanolic extract of soqatra island in experimental animals. *Univ. J. Pharm. Res.*, 2018, 3(5), 1-11.
15. Martysiak-Żurowska D, Wenta W. A comparison o ABTS and DPPH methods for assessing the total antioxidant capacity of human milk. *Acta Sci Pol Technol Aliment*. 2012;11(1):83-9.

16. Ramakrishna BS, Varghese R, Jayakumar S, Mathan M, Balasubramanian KA. Circulating antioxidants in ulcerative colitis and their relationship to disease severity and activity. *J Gastroenterol Hepatol.* 1997;12(7):490-4.
17. Taveira, Evolution of *Brassica rapa* var. *rapa* L. volatile composition by HS-SPME and GC/IT-MS, *Microchem. J.*, № 93, c. 140
18. Takuno, Phylogenetic relationships among cultivated types of *Brassicarapa* L. em. Metzg. as revealed by AFLP analysis, *Genet. Resour. Crop. Evol.*, № 54, c. 279
19. Sala, Anti-inflammatory and antioxidant properties of *Helichrysum italicum*, *J. Pharm. Pharmacol.*, № 54, c. 365
20. Cartea, Phenolic compounds in brassica vegetables, *Molecules*, № 16, c. 251
21. Wittkop, Status and perspectives of breeding for enhanced yield and quality of oilseed crops for Europe, *Euphytica*, № 170, c. 131
22. Valavala, Effect of mustard (*Brassica juncea*) leaf extract on streptozotocin-induced diabetic cataract in Wistar rats, *J. Food Biochem.*, № 35, c. 109
23. Kumar, Quality characteristics in rapeseed - mustard and role of some anti-nutritional factors in plant defense: future strategies, *J. Oilseed Brassica*, № 5, c. 87
24. Hill, Variation in glucosinolates in oriental Brassica vegetables, *J. Am. Soc. Hortic. Sci.*, № 112, c. 309
25. Rangkadilok, Developmental changes of sinigrin and glucoraphanin in three Brassica species (*Brassica nigra*, *Brassica juncea* and *Brassica oleracea* var. *italica*), *Sci. Hortic.*, № 96, c. 11
26. Gil, Benzylglucosinolate degradation in *Lepidium sativum*: effect of plant age and time of autolysis, *Phytochemistry*, № 19, c. 1365
27. Kessler, Plant responses to insect herbivory; the emerging molecular analysis, *Annu. Rev. Plant Biol.*, № 53, c. 299
28. Kesen, GC-MS-olfactometric characterization of the key aroma compounds in Turkish olive oils by application of the aroma extract dilution analysis, *Food Res. Int.*, № 54, c. 1987
29. Sales-Campos, An overview of the modulatory effects of oleic acid in health and disease, *Mini-Rev. Med. Chem.*, № 13, c. 201
30. Win, Oleic acid – the anti-breast cancer component in olive oil, *AU J. Tech.*, № 9, c. 75
31. Mahadkar, Gas chromatography mass spectroscopic (GCMS) analysis of some bioactive compounds from five medicinally relevant wild edible plants, *Asian J. Pharm. Clin. Res.*, № 6, c. 136
32. Manonmani, GC-MS analysis of bioactive components of an important medicinal fern *Actiniopteris radiata* (Swartz) link, *World J. Pharm. Res.*, № 4, c. 1860
33. Rimm EB, Ascherio A, Giovannucci E, Spiegelman D, Stampfer MJ, Willett WC. Vegetable, fruit, and cereal fiber intake and risk of coronary heart disease among men. *JAMA.* 1996;275(6):447-51.
34. Vanithadevi, B.; Anuradha, C.V. Effect of rosmarinic acid on insulin sensitivity, glyoxalase system and oxidative events in liver of fructose-fed mice. *Int. J. Diabetes Metab.*, 2008, 16, 35-44.
35. Sepici-Dincel, A.; Açıkgöz, S.; Çevik, C.; Sengelen, M.; Yeşilada, E. Effects of in vivo antioxidant enzyme activities of myrtle oil in normoglycaemic and alloxan diabetic rabbits. *J. Ethnopharmacol.*, 2007, 110(3), 498-503.
36. Pahwa, R.; Goyal, A.; Bansal, P.; Jialal, I. Chronic Inflammation. In *StatPearls*; StatPearls Publishing: Treasure Island, FL, USA, 2018.
37. Cash, J.L.; White, G.E.; Greaves, D.R. Chapter 17—Zymosan-Induced Peritonitis as a Simple Experimental System for the Study of Inflammation. *Methods Enzymol.* 2009, 461, 379–396.