

Determination of anxiolytic and antidepressant like activity of hydro alcoholic fruit extract of *Pyrus communis* (L.) and its impact on the memory after chronic dosing in animal models

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Abstract: The purpose of this study was to evaluate the anxiolytic and antidepressant activity of ethanolic fruit extract of *Pyrus communis* (pear), in comparison with escitalopram in rodents (rats and mice). Thirty Wistar rats of about 200-250gm and albino mice of 25-30gm, male gender were divided into three groups each comprising of (n=10) animal respectively. Control group received distilled water, positive control received 10mg escitalopram & treated group received 200mg/kg/day of *Pyrus communis* ethanolic fruit extract orally for 30 days. They were evaluated by using the open field test, forced swim test (FST), plus maze test, light and dark test, hole poking test, stationary rod test, water maze test & cage crossing activity. Results were expressed as mean \pm SD. Data was analyzed by using SPSS software (VERSION 21) one way ANOVA followed by Tukey test was used for post hoc analysis. Our result showed that fruit extract had significant antidepressant-like behavior in FST ($p < 0.001$), open field ($p < 0.05$), cage crossing ($p < 0.001$), significant anxiolytic activity in light and dark box test, plus-maze activity and significantly enhanced learning in water maze and stationary rod test when compared with control. The *Pyrus communis* fruit extract showed the anxiolytic and antidepressant-like profile in rats and mice. However, further studies need to be carried out in clinical trials for its use in different neuropsychological disorders.

Keywords: *Pyrus communis*, reactive oxidative species (ROS), reactive nitrogenous species (RNS), selective serotonin reuptake inhibitor (SSRI).

INTRODUCTION

Depression and anxiety disorders are devastating issues of the modern world. They are not same but have some common symptoms which can misguide the situation. The common symptoms experienced by the patients are insomnia, being nervous and irritable and feeling difficulty in concentrating. Both these disorders have different etiologies and behavioral symptoms. Sometimes chronic anxiety may cause depression (Sheehan *et al.*, 2017; Boyer, 2010). These disorders always take a significant place in clinical research. In depression and anxiety, the major part of amygdala and hippocampus undergoes hypotrophy and that is the cause they both occur together. Hypotrophy of both parts occur with long periods of depression (Irle *et al.*, 2010). Anxiety in simple words is the fright for any upcoming event in the future or any past awful experience which causes discomfort, fear and overreaction to a condition. On the other hand depression is dysphoria in our daily life and it is actually a disorder of mood.

High oxygen consumption by the neuronal cells and lipid rich environment makes the brain vulnerable to be affected, due to the formation of the reactive oxidative

species (Hulbert *et al.*, 2007). It is evident from many types of researches that an imbalance in the formation and detoxification of ROS in the brain, result in the occurrence of anxiety, depression, bipolar disorder, and schizophrenia (Salim, 2014). Use of antioxidants is the main approach to prevent or treat the patient suffering from the above-mentioned disorders (Bouayed, 2010). The antidepressant treatments include use of Selective Serotonin Reuptake Inhibitors (SSRI's). Although SSRI's are safer as compared to Tricyclic Antidepressant's but still few adverse effects limit its use, thus directing the scientist to discover safer medicine mostly from herbal origin.

Various plants possess medicinal properties as they are rich in phytonutrients that possess antioxidant property which may provide a novel approach for the prevention or treatment of above mentioned ailments. Natural antioxidants have minimal side effects as compared to synthetics with better therapeutic efficacy (Monika *et al.*, 2016). Polyphenols are naturally-occurring antioxidant substances which can have pharmacological actions on the CNS, which are the active constituent of herbs, fruits, vegetables. While fruits are reported to be high in vitamins, flavonoids etc and therefore are beneficial for the health (Zlotek *et al.*, 2015).

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Pyrus communis (*P. communis*) is the most common and highly cultivated specie (Kaur and Arya, 2012) all over the world. In Pakistan, different varieties of pear fruits are available, including Bagu-goshaa or Nakh, Nashpati and Bartlett etc (Manzoor *et al.*, 2013). Baluchistan, NWFP, Gilgit and different valleys of Baltistan are main regions of cultivation (Hussain *et al.*, 2013). *P. communis* contains high amounts of vitamins (A, B₁, B₂, B₃ and C) and minerals like Na⁺, K⁺, P⁺⁺, Ca⁺⁺, Mg⁺⁺, Zn⁺⁺ and Fe⁺⁺ (US department of agriculture, agricultural research service., 2018). Pear fruit contains pectin, carotenoids, anthocyanins, kaempferol, isorhamnetin, flavanols, quercetin, arbutin, rutin, epicatechin and plant sterols. The fruit also holds an extensive variety of phenolic acid for example chlorogenic acid, ursolic acid, coumaric acids, hydroxyphenolic acids (Li *et al.*, 2012; Kaur and Arya 2012; Hussain *et al.*, 2013; Ranjeet and Vikrant, 2012).

Pear fruits have various pharmacological properties like astringent, sedative, antioxidant, antibacterial, febrifuge (Kaur and Arya, 2012) and anti-inflammatory (Li *et al.*, 2011). Pear is best for diabetics and the obese persons because of high dietary fiber content in the fruit. Phenolic contents reduce the risk of cardiovascular diseases as well as used in the treatment of constipation as a laxative (Reiland & Slavin, 2015) and intestinal inflammation (Manzoor *et al.*, 2013). Arbutin decreases the melanin and is used as a human skin whitening agent. It also has some anti-inflammatory effects in leaves and barks (Kaur and Arya, 2012). Analgesic and spasmolytic effects has been reported in flowers (Velmurugan and Bhargava, 2013). Pears may also be useful in alcohol metabolism regulation (Reiland & Slavin, 2015).

The present study was designed to investigate the anxiolytic and anti-depressant effect of pear extract in rodents and to assess its effect on memory too.

MATERIALS AND METHODS

Preparation of crude extract

P. Communis fruit was purchased from the local market of Karachi. It was identified in the Department of Pharmacognosy, Faculty of Pharmacy, Institute of Pharmaceutical Sciences, Jinnah Sindh Medical University, Karachi Pakistan. In preparation of extract, whole fruits were cut into small pieces and macerated in a hydro alcoholic solution (water/methanol; 30/70) for 15 days at room temperature. The hydro alcoholic solution was strained, the solvent was then evaporated under reduced pressure through the rotatory evaporator to obtain the crude extract of *P. Communis* fruit. The extract was then stored in refrigerator for further use.

Phytochemical analysis

The extract of *P. Communis* was screened for various plant constituents like alkaloids, saponins, carbohydrate and flavonoids, etc. The methods employed for the

determination of plant constituents were chosen, as executed by previous workers (Ly *et al.*, 2021). Various test were performed for example, molisch test, fehling test, benedicts test, moore test, Dragondraff's test, salkowski test, Foam test and lead acetate test etc.

Determination of flavonoidal content

After extraction procedure, identification and determination of total flavonoid contents was determined at different concentration as reported by the Imam and coworkers (2017) which confirmed the presence of flavonoids.

Antioxidant activity of *Pyrus Communis*

The antioxidant activity of *P. communis* extract was analyzed using DPPH free radical scavenging and metal chelating assay besides that total phenolic content was also evaluated (Ly *et al.*, 2021).

Animals

Long-term oral administrations of test extract and standard drug was done in Wistar rats. The range of average weight of selected rodents was from 200g to 250 gm for rats and 25-30gm for mice. All animals were arbitrarily grouped into three groups, each group having ten animals.

1st group of animals = control group distilled water was administered.

The 2nd group of animals=treated group for standard drug escitalopram (10mg adult dose) (Li *et al.*, 2017)

While 3rd group =treated group for the hydroalcoholic extract of *P. communis* fruit for CNS activities.

Housing

The room temperature was kept in the range from 20 to 24° C and relative humidity 25±5%, with dark/ light cycle at the Department of Pharmacology, Institute of Pharmaceutical Sciences, Jinnah Sindh Medical University, Karachi Pakistan. Animals were provided water and food ad libitum. Two animals per cage were housed under regular circumstances for 1 week before the start of treatment. All pharmacological experiments were performed in accordance with NIH (National Institutes of Health) guidelines on Laboratory Animal Care (NIH publication #85-23).

Dose calculation

As per Velmurugan and Bhargava, (2013) the lethal dose of *P. Communis* is 2000mg/kg. So the effective dose ED 50 was computing by using the formula given below:

$$\begin{aligned}LD50 &= 2000\text{mg/kg} \\ ED50 &= LD50/10 \\ &= 2000/10 \\ &= 200 \text{ mg/kg}\end{aligned}$$

The distilled water was given to control group orally in the same manner as standard drug and test extract were administered to the treated groups.

General behavioral activities

Behavioral despair test

Behavioral despair test (force swim test) is generally used tool to induce depression in rodents. An acrylic glass cylinder (30cm; height, 12cm; diameters) filled with water ($24\pm 2^{\circ}\text{C}$) at the specific level (24cm height) is used as a apparatus. At 15th and 30th days of treatment, the swimming duration was noted (Drugan *et al.*, 2010). All groups of albino rats were forced to swim individually. The water of the apparatus was changed after every round to help prevent bacterial contaminations (Crawley, 2007).

Home cage activity

The apparatus consists of square shaped acrylic glass cages with a transparent floor. The number of exploration was recorded for 10 minutes after placing rats independently in the enclosures (Najam and Anser, 2011). Exploratory activity was tested at the 15th and 30th day of dosing for all groups.

Open field test

This test is an important tool for the evaluation of anxiety, examination, and impulsive locomotion. The open field box was constructed with acrylic material having a transparent floor with 76cm x 76cm x 40cm dimensions. The floor had 16 squares of 18x18 cm size (Seibenhener & Wooten, 2015). Albino rats were placed in the corner facing towards the wall. The total amounts of entries in the squares were calculated for 10 minutes. All animal groups were evaluated at day 15th and day 30th (Tamada *et al.*, 2010).

Light and dark test

Light and dark test is the most important test for anxiolytic activity of drugs (Bourin & Hascoët, 2003). The apparatus has the standard size in which one third part of the compartment is dark and the two thirds of the compartment has light with an exterior size of 46 x 27 x 30 cm (l x b x h). The rats were located in the illuminated area (700 lux). The modified apparatus was used as stated by Khan and Haleem (2008). The rodents preferred dark compartment because of their habit and habitat. Treated animals (rats) were familiarized to the light compartment with their back the opening as by Peng *et al* (2000). The time spent in light area and the number of shifts among the two compartments were recorded for 10 minutes in all the groups (Bourin and Hascoet, 2003). The percent duration in the light compartment (Time in light compartment/ Total time x 100), was measured.

Hole poking test

The hole poking test is used for the analysis of anxiogenic, anxiolytic and exploratory activity of mice. The apparatus used was made up of wooden material (60x30cm) with evenly spaced 16 holes. Each mouse was placed on the corner and permitted to discover the arena. The total number of hole pokings were counted over the

period of 10 minutes. The curiosity of hole poking is indicative of degree of anxiousness and explorations. All groups of mice were assessed after 15 and 30 days of dosings (Pandey *et al.*, 2016).

Elevated plus maze

This test is used for the analysis of anxiogenic and anxiolytic agents in rodents. After 15 and 30 days of dosing, the mice were evaluated for five minutes on an elevated-plus maze as per the procedure described by Pandey and co-workers (Pandey *et al.*, 2016). The mouse was placed at the intersection between the open and closed arms, the time spent in the open arms was recorded after 15 days and 30 days of dosing.

Water maze test

The test was performed to evaluate the cognition process of the rodents (D'Hooge and De Deyn, 2001). The apparatus consist of a round container filled with water at room temperature. The maze depth is about 3 feet and its diameter is 6 feet. The starch was dissolved in water in order to make water opaque so the rat is unable to see the platform. Water level is kept 2cm and platform is placed fixed throughout the procedure (Joseph Nunez, 2008).

The rats were trained by placing in the water tank, facing towards the wall of the tank and allowed 120 seconds to locate and climb onto the hidden platform. The rats were allowed to stay on the platform for 10 seconds. If it failed to find the platform within the allowed time it was guided gently onto the platform. The memory function of rat was tested by recording the retention latency (RL; the time taken by each rat to locate the hidden platform 1 h (short term) 24h (long term) after training).

Stationary rod test

Stationary rod test is a common test which is used to assess capability of learning. It consists of stainless steel rods and both ends contain platforms. Albino mice were trained daily on this apparatus (Kishioka *et al.*, 2009). Assessment of the extract and drug was done by evaluating the learning and memory of both the groups i.e treated and control. The procedure was adopted as stated by Humera *et al.*, 2014. The test was repeated after 15 and 30 days administering water, test and standard drugs.

STATISTICAL ANALYSIS

The animal's behavioral activities were statistically analyzed by one way ANOVA followed by Tukey post hoc test of multiple comparisons using SPSS version 21. The results were expressed as mean \pm SEM and were considered statistically significant at $p\leq 0.05$ and highly significant at $p\leq 0.001$. Paired T-test was also performed in test group.

RESULTS

Table 1 and 2 show the phytochemical screening, Flavonoidal content and anti-oxidant activity of the pear extract. Fig 2 displayed the data analysis done by using one way ANOVA it showed significant results in home cage crossings, the difference within groups and in between group ($F=34.7$, $p \leq 0.001$) and ($F=58.38$, $p \leq 0.001$) after 15th and 30th days of dosing of respectively. Tukey test for post hoc analysis indicated that the group treated with *Pyrus communis* revealed an enhanced number in cage crossings after 30th day of administering ($p \leq 0.05$) when equated with control group and significantly decreased when compared with escitalopram. The pair t test analysis of the data showed a highly significant increase in cage crossings after 30th day of dosing when compared with after 15th day of administration ($p < 0.001$).



Fig. 1: Fruit of *Pyrus communis* (Ranjeet and Vikrant, 2012).

Fig. 3 revealed momentous results in forced swimming test among the groups and in the groups ($F=54.07$, $p \leq 0.001$) and ($F=35.07$, $p \leq 0.001$) respectively after 15th and 30th days of dosing. Tukey test for post hoc analysis revealed major rise in swimming time after 15th as well as after 30th days of dosing in the treated group in link with control ($p \leq 0.001$), the difference was insignificant when compared with positive control (escitalopram). The pair t test analysis of the data showed a major rise in swimming time after 30th day of dosing when compared with after 15th day of administration ($p \leq 0.001$).

Fig. 4 revealed momentous differences in open field activity among groups and within groups ($F=12.65$, $p \leq 0.001$) and ($F=11.47$, $p \leq 0.001$) respectively after 15th and 30th day of administering. Tukey test for post hoc revealed a great decrease in overall quantity of crossings after 15th day ($p < 0.001$) but a moderately significant increase in total number of crossings after 30th days of administering ($p < 0.01$) when related with control. The difference was insignificant when compared with treated on 30th day. The

paired t test analysis of the data revealed a vast rise in total crossings in open field after 30th day of administering when related with after 15th day of administration ($p \leq 0.001$).

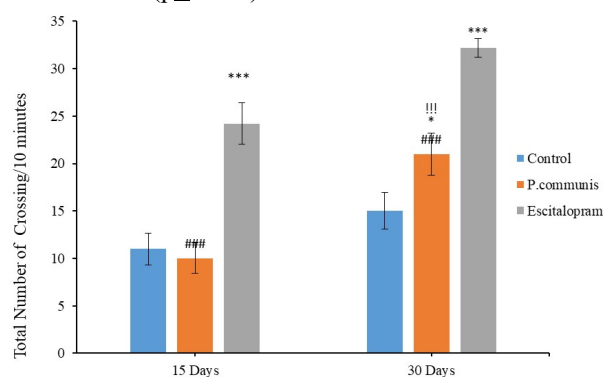


Fig. 2: Effect of ethanolic extract of *Pyrus communis* fruit on cage crossing test

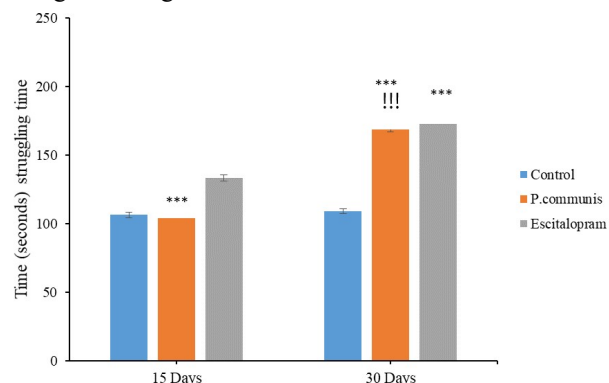


Fig. 3: Effect of ethanolic extract of *Pyrus communis* fruit on forced swim test

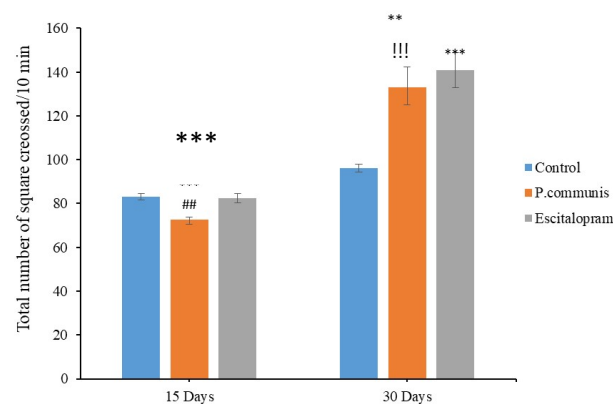


Fig. 4: Effect of ethanolic extract of *Pyrus communis* fruit on open field test.

Fig.5 revealed major difference in duration spent in open arm of plus maze among groups and in the groups ($F=9.3$, $p \leq 0.001$) and ($F=15.63$, $p \leq 0.001$) respectively after 15th and 30th day of treating. Turkey's test of Post hoc analysis revealed that animals given crude extract of *Pyrus*

communis highly significantly increased the time duration in open arm of plus maze ($p \leq 0.001$) after 15th day and after 30th days of dosing. On 30th day the time spent in open arm was highly significantly increased as compared to escitalopram. The paired t test analysis of the data showed a significant increase in time spent in open arm of plus maze after 30th day of treating when related with after 15th day ($p \leq 0.05$).

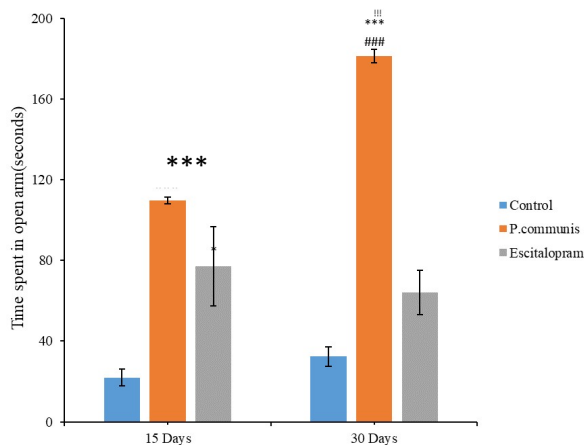


Fig. 5: Effect of ethanolic extract of *Pyrus communis* fruit on the time spent in open arm of elevated Plus maze.

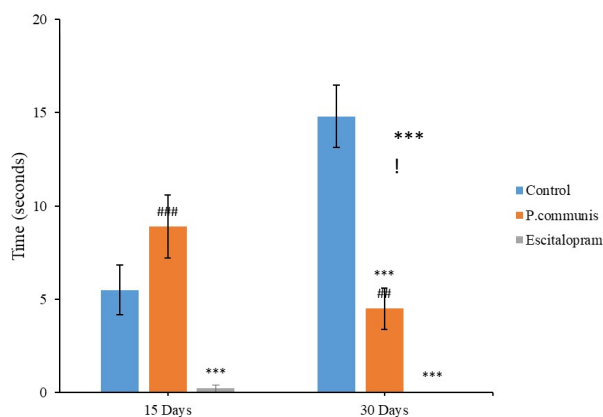


Fig. 6: Effect of ethanolic extract of *Pyrus communis* fruit on Water maze test.

Fig. 6 indicated major difference in water maze test among groups and in groups ($F=17.59$, $p \leq 0.001$) and ($F=42.79$, $p \leq 0.001$) respectively after 15th and 30th day of treatment. Tukey test post hoc analysis revealed that groups administered with crude extract of *Pyrus communis* showed no significance in time taken to reach the platform after 15th day while highly significant decreased time was taken after 30th days of dosing ($p \leq 0.001$). Escitalopram showed significant reduction in time taken to reach the platform when compared with treated. The paired t test analysis of the data showed an insignificant drop in interval to get the platform in water maze after 30th day of dosing when related with after 15th day of administration.

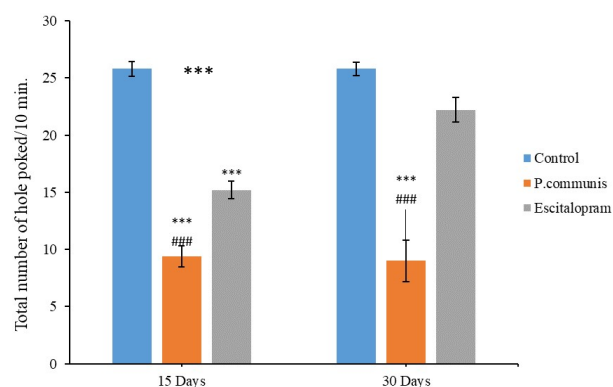


Fig. 7: Effect of ethanolic extract of *Pyrus communis* fruit on Hole Poking test.

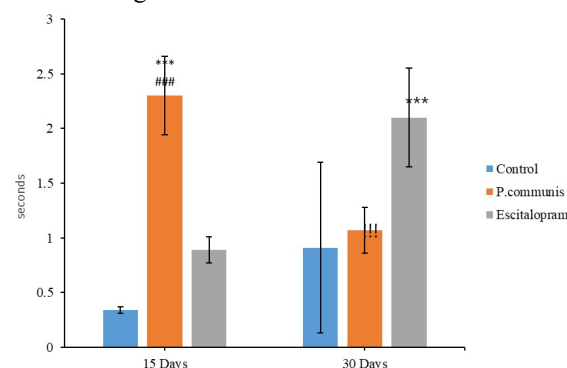


Fig. 8: Effect of ethanolic extract of *Pyrus communis* fruit on stationary rod test.

Fig.7 indicated important difference in hole poking test among groups ($F=111.415$, $p \leq 0.001$) and ($F=49.45$, $p \leq 0.001$) respectively after 15th and 30th day of administration. Tukey test for post hoc analysis revealed that group administered with crude extract of *Pyrus Communis* highly significantly decreased the hole poking activity ($p \leq 0.001$) after 15th day as well as after 30th days of dosing. Escitalopram showed significant poking when compared with treated throughout the study. Although the pair t test analysis of the data showed a significant increase in hole pokings on 30th day of dosing when linked with after 15th day of administration ($p \leq 0.05$).

Fig.8 showed a significant difference in stationary rod test amongst and within groups ($F=23.28$, $p \leq 0.001$) and ($F=1.64$, $p \leq 0.001$) respectively after 15th and 30th day of treatment. Tukey test after post hoc analysis revealed that group treated with crude extract of *Pyrus communis* took highly significantly increased period to reach to the platform of stationary rod after 15th day ($p \leq 0.001$) while the difference was insignificant on 30th day of dosing. The time taken by escitalopram was increased on 30th day as compared to control. The pair t test analysis of the data showed an moderately major decline in time to touch the platform on stationary rod apparatus after 30th day of

dosing when linked by after 15th day of administration ($p < 0.01$).

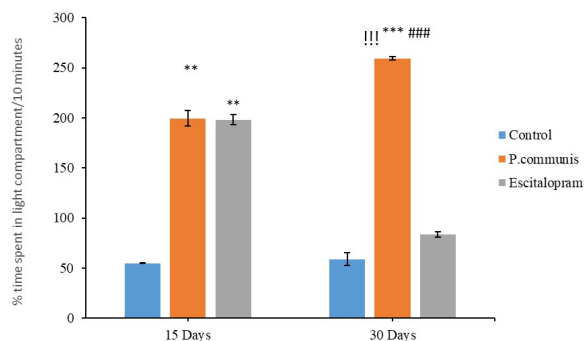


Fig 9: Effect of ethanolic extract of *Pyrus communis* fruit on light and dark test

One-way ANOVA followed by Tukey test for multiple comparison was applied where *, ** and *** showed significant, moderately significant and highly significant difference as compared to control. #, ## and ### display significant, moderately significant and highly significant difference when treated was compared to escitalopram (positive control). !,!! and !!! display significant, moderately significant and highly significant difference as compared to one month (30 days) values.

Fig.9 revealed major difference in light and dark test among groups and within groups ($F=7.3$, $p \leq 0.01$) and ($F=87.14$, $p < 0.001$) respectively after 15th and 30th day of administration. Tukey test for post hoc analysis revealed that group treated with crude extract of *Pyrus Communis* moderately significantly increased the %time spent in light compartment ($p \leq 0.01$) after 15th day and vastly enhanced it after 30th days of administration ($p < 0.001$). Escitalopram showed highly significantly reduced time spent in light area as compared to treated on 30th day. Although the pair t test analysis of the data showed a major rise in % time consumed in light box after 30th day of dosing when correlated with after 15th day of treating ($p \leq 0.05$).

DISCUSSION

Our result showed that *Pyrus communis* fruit extract

Table 1: Phytochemical Analysis

Plant/part used	Carbohydrates	Flavonoids	Alkaloids	Tannins	Saponins
<i>P. communis</i> /Fruit	++	++	+	++	+

Where + shows mild presence and ++ shows moderate presence

Table 2: Flavonoidal content and antioxidant activity of *Pyrus communis*

Plant extract	Total phenolic content (mg GA/g dry extract)	DPPH free radical scavenging capacity (1mg/ml)	Chelating activity of 1mg/ml extract	Flavonoidal content QE ($\mu\text{g/g}$)
<i>Pyrus communis</i> (fruit extract)	7.05	9.5	5.05	11.37+ 3.4

GA=gallic acid, QE= quercetin, DPPH=1,1-Diphenyl-2-picrylhydrazyl radical

induces significant antidepressant-like behavior in FST ($p < 0.001$), open field ($p < 0.05$), cage crossing ($p < 0.001$) when compared with control. The anxiolytic activity was observed in hole poking activity ($p < 0.001$), light and dark box test ($p < 0.05$) and elevated plus maze test ($p < 0.001$) after 15 and 30 days of dosing. While the memory of the rodent was assessed with the help of water maze test ($p < 0.05$) and stationary rod test ($p < 0.001$) after 30 days of dosing.

Many plants are reported to reduce oxidative stress by decreasing the production of ROS and RNS. *Pyrus communis* possesses a number of potent ingredients that are antioxidant in nature. Among them are polyphenols, flavonoids glycosides (quercetin, iso-quercetin, kaempferol) carbohydrates (pectin, sucrose), various ions sodium ion, Phosphorous ion, magnesium ion, zinc ion, various vitamins (vitamin A, C, B Complex) etc. Zhang *et al* (2011) had reported that quercetin decreases NO and ROS production.

The studies done by Machado *et al.* (2008) and Guan and Liu, (2016) support our study that diglycoside flavonoid rutin may increase adrenaline and serotonin availability. Thus proving that the *P. communis* plays a significant role in the elevation of neurotransmitter and be helpful in the alleviation of depression. Antioxidants produce their reducing effects through transfer of hydrogen atom, or electron transfer or metal chelation.

Cage crossing activity was found increased after chronic dosing which showed an improvement in locomotion and exploration which can be related to the presence of flavonoidal and mineral content of *P. communis*.

The swimming time was found increased after administration of *P. Communis extract* as compared to control rats. The study done by Bhutada and coworkers (2010) confirmed that quercetin rich fruits, showed antidepressant like activity as it decreases the expression of Corticotrophin releasing factor (CRF) in brain, thus decreasing the immobility time. It was also reported by Liaquat *et al.* (2018) that repeated use of quercetin rich fruits improved the cholinergic and serotonergic

functions. Oxidative stress-mediated cell death occurs due to elevated levels of Ca^{++} ion concentration that results in increased energy expenditure and subsequent initiation of cyto-skeletal degradation ultimately resulting in neuronal loss and leading towards depression. Studies done by Sankanashi (2008) had reported that quercetin is capable of decreasing the increased levels of Ca^{++} ion concentration that ultimately is the reason of its antidepressant effect. The increase in swimming time can also be related to a rise in magnesium amounts after administration of *P. communis* which is reported to be rich in magnesium, zinc etc. Increase in the levels of magnesium produces antidepressant like activity and is proved for the relieve of etiology of depression (Boyle, Lawton, & Dye, 2017).

The rodents always prefer staying in dark regions, away from the light and decreased movement in response to any aversive stimuli. This study showed that duration of spending time in light box is remarkably increased which is supporting the anxiolytic like activity of the ethanolic extract of *P. communis*. Li and his coworkers (2016) had reported in their study that quercetin modulates the 5HT levels, which after chronic administration modulates the 5HT and noradrenaline levels. The Elevated Plus Maze is considered a valid animal model of anxiety because the animal become stressed when exposed to novel open spaces and the fear of height while moving through narrow and elevated spaces which induces anxiety in them. The total time spent in the open arm of plus maze was increased after oral administration of *P. communis* extract. Our observation is supported by the studies done by Samad and his coworkers (2018) who had confirmed that quercetin inhibit MAO enzyme presynaptically leading to increase levels of 5HT in the synaptic cleft ultimately increasing the availability which helps in alleviation of depression and anxiety and increases locomotion. Another literature supporting our study states that the bioflavonoid quercetin improves the time spent in the light compartment in light and dark box, it also improves time spent in open arm of plus maze with little effect on locomotion.

This effect is attributed to modulation of 5HT levels through 5HT1A receptor activation (Li et al., 2016) that in turn produces anxiolytic as well as antidepressant-like effect of ethanolic extract of *P. communis*. Li et al (2016) also confirmed that chronic administration will elevate the level of 5HT (which were decreased in depression) through down regulation of 5HT1A receptor.

After chronic administration of *P. communis* the exploration in the open field were increased, that showed an improvement in locomotion and exploration which shows reduction in anxiety and depression. This effect can be attributed to effect of quercetin and kaempferol which are present abundantly in *P. communis*. Both of

these bioflavonoids increase the levels of catecholamines in the brain of rodents (Yan et al., 2015).

The hole poking test helps in the assessment of stress induced behavioral reaction in animal to novel environments. When the animal is exposed to novel environment anxiety and depression may be induced. The total number of hole poking were found to be decreased in treated group of mice in comparison with control showing anxiolytic like effect, the animal was fearless and more time was spent in exploring the area rather than poking the hole. This can be attributed to presence of minerals and polyphenolic compounds for example magnesium, potassium, iron, quercetin, kaempferol etc (after administration of *P.communis* extract), which is reported to increase GABAergic activity via decreasing the glutamate activity (Vink & Nechifor (2011).

The Morris water maze apparatus is used in ethological studies for the determination of effect of the drugs on memory. Its activity states the time required to discover the platform which was unseen because of opaque water, which may reveal the effect of the test agent on short term as well as long term memory after acquisition of training and reconsolidation (Liaquat et al., 2018). In our study after administration of ethanolic extract of *P.communis* memory was improved after chronic dosing. To assess the learning and motor coordination, the most common test is stationary rod test. The improvement was shown in animal after the chronic dosing which clearly indicated the variation of cholinergic transmission (Samad et al., 2018). The memory enhancement may possibly be due to bioflavonoids like quercetin and kaempferol, it increases the level of hippocampal 5HT and norepinephrine through its monoamine oxidase enzyme inhibition (Bandaruk et al.,2012) and it is also related to its neuroprotective action by scavenging free radical oxygen specie (generated due to stress) (Marcer et al., 2005; Sharma et al.,2015; Samad et al.,2018). Various studies have been done which showed an increase in levels of antioxidant enzyme for eg. SOD, catalase, Glutathione peroxidase after administration of quercetin (Yoshino et al. 2011; Kumar and Goyal 2008). Beside increasing the levels of 5HT quercetin is also reported to increase the levels of acetyl choline (Samad et al., 2018). through acetyl cholinesterase enzyme inhibition (Islam et al., 2013). which improves the cognition and learning in stressed animal (Mohammadi et al., 2014).

CONCLUSION

We have made an effort to evaluate the antidepressant and anxiolytic like activity of *P. communis*. The study showed potential of *P. communis* to produce antidepressant and anxiolytic like activity. Besides that it has an additional benefit of enhancing the memory which is usually not present in anti-depressant and anxiolytic drugs. Its phytoconstituents are antioxidant in nature and could be

effective in prevention and relief of anxiety and depression. Since exact mechanism is not known. Therefore further studies at molecular level are suggested in future.

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