CHAPTER- IX

ANTI-ACID SECRETORY AND ANTIULCER ROLE OF AQUEOUS EXTRACT OF RIPE FRUIT’S PULP OF *Cucurbita pepo* Linn. AND RANITIDINE AGAINST CEREBELLAR NODULAR LESION INDUCED GASTRIC ULCERATION BY ALTERING pH, VOLUME OF SECRETION, ACID SECRETION AND MUCUS CONTENT

Several neuro-physiological defective causations of central nervous system (CNS) such as cerebellar nodular lesion (CNL) and hypothalamic dysfunction as well as several neuro-physiological defective causations of enteric nervous system (ENS) hamper normal physiology of gastrointestinal tract (GIT). Peptic ulcer disease (PUD) is influenced by defective cerebellum. Severe ulceration (Sarkar *et al.*, 2006) caused by numerous defective functions of gastrointestinal tract (GIT) such as increased volume of secretion, excessive acid secretion (Wolfe, 1969), excessive acid-peptic activity, hyper-motility of gastrointestinal tract (GIT) (Voronin, 1938) and reduced mucus secretion etc due to complex neurogenic neuropathophysiological conditions of higher nervous system.

Cerebellum has been claimed to modulate various autonomic and visceral physiological functions (Ito, 1978). Lesions of restricted area of cerebellum produce gastric hyper-secretion and ulceration (Wolfe, 1969). Posterior cerebellar lesions induce severe focal inflammatory ulcers at the stomach (Wolfe, 1969; Brooks, 1967) which is associated with extensive damage of the surface epithelium cells, leading to necrotic ulcers with a significant reduction of mucus content (total CHO: P), pH and significant increase in volume of secretion, excessive acid secretion, excessive acid-peptic activity of gastric juice etc (Sonnenberg, 1995; Sarkar *et al.*, 2006; Guha *et al.*, 2002; Sarkar and Guha 1999; Maiti and Guha, 1978).

The controlled secretion of hydrochloric acid (HCl) under normal physiological condition does not cause any gastric problems but sustained excessive secretion is a
grave pathological condition, which is termed as hyperchlorhydria. Therefore, various adverse physiological conditions as well as intake of several drugs such as non-steroidal anti-inflammatory drugs (NSAIDs; aspirin, indomethacin, ibuprofen and phenylbutazone etc), they are commonly used as painkillers, cause excessive acid secretion with a significant reduction of mucus content (total CHO: P) which leads to peptic ulcer disease (PUD).

The anti-acid secretory and antiulcerogenic plant extracts, composites, preparations, formulations of licorice root (Goso et al., 1996), calendula, chamomile (Beil et al., 1995), garlic (Sivam et al., 1997), thyme (Ernst, 1999), cinnamon (Graham et al., 1999), corydalis (Chang et al., 1986), comfrey (Graham, 1999), *Azadirachta indica* (Garg et al, 1993), *Stachydrpheta cayennensis* (Vela et al., 1997), *Cistus laurifolias* L (Yasilada et al, 1997), *Musa paradisiacal* (Goel and Bhattacharya, 1991) and phytochemicals which are currently used as herbal remedies that can effectively control these conditions.

*Cucurbita pepo* Linn. (Cucurbitaceae) (Paris et al., 2003; Paris, 2009) plant, commonly known as pumpkin, is available throughout India and consumed as vegetable (Sarkar and Guha, 2008; Koike et al., 2005; Mongkolsilp et al., 2004; Sammon et al., 2003) and medicinal food (Hilgert and Gil, 2007) in various parts of the world. The plant is used to prepare infants food formulations (Ezeji and Ojimeukwe, 1993) for the improvements in protein quality (Gibson et al., 2006). Different parts of *Cucurbita pepo* Linn. (*C. pepo*) have been used as traditional medicine in Ayurveda (Caili et al., 2006). This plant is used to treat cuts, injuries and swellings (Lans, 2007). Plant is also used as anthelmintics (Al-Qura’n, 2008; Lans et al., 2007), diuretic (Al-Qura’n, 2008) and used to expel tapeworms from intestine (Lans et al., 2006).

Fruits of *Cucurbita pepo* Linn. (Cucurbitaceae) (Paris et al., 2003; Paris, 2009) are used as vegetable (Sunilson et al., 2009; Sarkar and Guha, 2008; Koike et al., 2005; Mongkolsilp et al., 2004; Sammon et al., 2003; Linskens and Jorde, 1997) and fruit’s pulp of pumpkin is used in dyspepsia and enteritis (Orlandelli, 1951), intestinal diseases (Francois et al., 2006). Fruits are used to relieve inflammation (Caili et al., 2006). It has been reported that pumpkin is consumed as a diet to increase the pH of fasting gastric sample (Sammon et al., 2003) and the dietetic management of patients undergoing
gastric operations is also carried out by supplementation of pumpkin (Loranskaia et al., 1986). Fruits are applied to treat liver disorder (Sezik et al., 2004).

Several antiulcer compounds such as terpenoids and triterpenoids (Wang et al., 2008), glycosides such as cucurbitacin B, cucurbitacin D, cucurbitacin E, cucurbitacin F (Feng et al., 2007), cucurbitacin L and cucurbitacin K (Wang et al., 2007), sulfhydryls like glutathione (Alosi et al., 1988), cysteine (Cys or C) (Fahmy et al., 2008; Pham et al., 1985; Allen, 1979; Kleinig et al., 1975; Ogura et al., 1972; Walker, 1972; Kataoka et al., 1998), S-adenosyl methionine (SAM) (Huang et al., 1991), methionine (Met or M) (Yoshida et al., 2005); phenolic compounds such as tannin (Ojiako and Igwe, 2008; Silveira et al., 1996), syringic acid (Dragovic-Uzelac et al., 2005), phenolic phytochemicals (Kwon et al., 2007), polyphenol and phenolic contents (Mongkolsilp et al., 2004), polyamines (Nishimura et al., 2006; Martinez-Tellez et al., 2002), betacarotene (Veda et al., 2006), pro-vitamin A carotenoids (Azevedo-Meleiro et al., 2007; Priyadarshani and Chandrika, 2007; Seo et al., 2005; Mongkolsilp et al., 2004; Manzi et al., 2002; Gonzalez, 2001; Rodriguez-Amaya, 1999; Kune et al., 1992; Arima and Rodriguez-Amaya, 1990; Arima and Rodriguez-Amaya, 1988), vitamins A (Lans et al., 2007; Ahmed et al., 2003; Ribaya-Mercado et al., 1999), vitamin C (Hancock et al., 2008; Mongkolsilp et al., 2004; Hancock et al., 2003) and vitamin E (Franke et al., 2007; Tadmor et al., 2005; Imaeda et al., 1999; Sedghi et al., 2008) are present in C. pepo fruit have been reported. Furthermore, some antioxidant-type elements such as selenium (Se) (Yoshida et al., 2005; Stibilj et al., 2004), Zinc (Zn) (Fan et al., 2006) and cadmium (Cd) (Qadir et al., 2000) have been found in C. pepo fruit.

Thus the present study was undertaken to determine the anti-secretory and antiulcer activity of aqueous extract of ripe fruit’s pulp C. pepo against cerebellar nodular lesion (CNL) induced gastric and duodenal ulceration in rat model.

Materials and methods:

Animal grouping and treatment:

Thirty (30) rats were divided into five groups of 6 rats each. Group I animals comprised control group. Group II was C. pepo extract treated, Group III was cerebellar nodular lesion (CNL), Group IV was C. pepo pretreated and cerebellar nodular lesion
The dry extract was dissolved in distilled water. Group I and Group III rats were given distilled water with approximately same volume of C. pepo extract orally by orogastric cannula. Group II and group IV rats were pretreated with selected dose of C. pepo extract (400 mg/kg body weight) and group V rats were pretreated with selected dose of ranitidine (10 mg/kg body weight) orally by orogastric cannula once daily for 14 consecutive days at a particular time (10:30-11:30 hrs) in every day. On 8th day, group III, group IV and group V rats were anaesthetized with sodium pentobarbital (40 mg/kg body weight; Abbott India Ltd) intraperitoneally (i.p). Cerebellar nodular lesion (CNL) was performed as per stereotaxic co-ordinates (AP=12.8, L=0.4, D=6.8)ellegrino and Cushman, 1967). Electrolytic lesions were made in the nodular cerebellum by conventional bipolar electrode (insulated by epoxylite with 0.5 mm tip exposed) using 1.5 mA DC (milli Ampere Direct Current) for 20 seconds for consecutive 3 days.

Gastric cannulation and gastric juice collection:

Prior to implant of gastric cannula, all rats were kept overnight fasting, but they were allowed free access to water. Stainless steel gastric cannula was implanted under anaesthesia according to the method of Guha et al., 1974. After post operative period of 7 days, rats were fasted for overnight but they were allowed free access to water and placed in the restraining cages (No. 51339, Stolting Co, Chicago). Prior to gastric juice collection stomach(s) were lavaged with 0.9% saline until the efferent was clear of any food particles. Gastric juice was collected through sialistic tube firmly attached with the cannula by opening the screw (Guha et al., 1974).

Parameters studied:

(I). Acidity: Acidity was measured with 0.01 (N) NaOH solution by using 0.5% Topper’s reagent and 1% phenolphthalein as an indicator (Guha and Maiti, 1990).

(II). Estimation of mucus content: 0.5 ml of gastric juice together with 5 ml of absolute alcohol was centrifuged for 20 minutes. The precipitate obtained was dissolved in 0.5 ml of 0.1 (N) NaOH solutions. Then 0.1 ml from this solution was taken for estimation of total protein and rest 0.4 ml was hydrolyzed for estimation of total hexose, hexosamine, sialic acid and fucose.
(i). **Estimation of total protein:** 0.1 ml of unhydrolyzed precipitate was taken for estimation of total protein spectrophotometrically (Lowry et al., 1951).

(ii). **Estimation of total hexose:** 1 ml of hydrolyzed solution was taken for estimation of total hexose spectrophotometrically (Elson and Morgan, 1953).

(iii). **Estimation of hexosamine:** 1 ml of hydrolyzed solution was taken for estimation of hexosamine spectrophotometrically (Dische and Borenfreund, 1950).

(iv). **Estimation of fucose:** 0.5 ml of hydrolyzed solution was taken for estimation of fucose spectrophotometrically (Dische and Shettes, 1948).

(v). **Estimation of sialic acid:** 1 ml of hydrolyzed solution was taken for estimation of sialic acid spectrophotometrically (Warren, 1959).

(III). **Histochemical study:** The mucus of stomach and duodenal tissues were stained with combined Alcian-blue-PAS stain (Mowry, 1956).

**Results:**

Result showed that treatment with aqueous extract of ripe fruit’s pulp of *C. pepo* (400 mg/kg body weight) decreased acid output, increased the total carbohydrate (e.g. hexose, hexosamine, fucose and sialic acid) and decreased protein content followed by increased mucus (CHO: P) secretion as compared to control rats. After cerebellar nodular lesion (CNL), ulceration was produced by increased acid output interfering the mucosal defensive factors (as evidenced by decreased carbohydrate and decreased protein content) followed by decreased mucus (CHO: P) secretion. But pretreatment with aqueous extract of ripe fruit’s pulp of *C. pepo* (400 mg/kg body weight) and ranitidine (10 mg/kg body weight) exhibited protective effect by increasing the mucus (CHO: P) secretion as evidenced by increased total carbohydrate (e.g. hexose, hexosamine, fucose and sialic acid) and increased protein content and thereby reduced the corrosive effect of gastric acid (Table 9.1).
Table 9.1: Effect of ripe fruit's pulp extract of *C. pepo* (400 mg/kg body weight) and ranitidine (10 mg/kg) on gastric secretory factors (pH, volume of secretion, acid output, hexose, hexosamine, fucose, sialic acid, total carbohydrate, total protein and mucus; carbohydrate protein ratio; CHO: P) against cerebellar nodular lesion (CNL) induced ulcer model.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>pH</th>
<th>Volume of secretion (ml/hr)</th>
<th>Acid output (μEq/L/hour)</th>
<th>Hexose (μg/ml)</th>
<th>Hexosamine (μg/ml)</th>
<th>Fucose (μg/ml)</th>
<th>Sialic acid (μg/ml)</th>
<th>Total carbohydrate (CHO) (μg/ml)</th>
<th>Total protein (μg/ml)</th>
<th>Carbohydrate-Protein (CHO: P)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>2.30</td>
<td>1.175</td>
<td>38.55</td>
<td>318.40</td>
<td>183.96</td>
<td>126.01</td>
<td>86.183</td>
<td>714.56</td>
<td>344.11</td>
<td>2.07</td>
</tr>
<tr>
<td>±0.14</td>
<td>±0.061</td>
<td>±2.199</td>
<td>±0.106</td>
<td>±1.91</td>
<td>±1.682</td>
<td>±0.94</td>
<td>±4.3232</td>
<td>±5.633</td>
<td>±0.02</td>
<td></td>
</tr>
<tr>
<td><em>C. pepo</em></td>
<td>4.00</td>
<td>0.745</td>
<td>28.33</td>
<td>437.50</td>
<td>288.00</td>
<td>156.16</td>
<td>114.66</td>
<td>996.33</td>
<td>312.00</td>
<td>3.193</td>
</tr>
<tr>
<td>±0.36</td>
<td>±0.007</td>
<td>±1.236</td>
<td>±2.291</td>
<td>±1.15</td>
<td>±1.166</td>
<td>±2.201</td>
<td>±2.94</td>
<td>±1.064</td>
<td>±0.115</td>
<td></td>
</tr>
<tr>
<td>CNL</td>
<td>1.58</td>
<td>2.20</td>
<td>64.46</td>
<td>186.08</td>
<td>134.6</td>
<td>57.71</td>
<td>44.45</td>
<td>422.85</td>
<td>264.65</td>
<td>1.597</td>
</tr>
<tr>
<td>±0.2*</td>
<td>±0.15*</td>
<td>±3.188*</td>
<td>±1.15*</td>
<td>±1.905*</td>
<td>±2.56*</td>
<td>±2.32*</td>
<td>±4.9255*</td>
<td>±2.999*</td>
<td>±0.009*</td>
<td></td>
</tr>
<tr>
<td><em>C. pepo</em> +CNL</td>
<td>2.75</td>
<td>0.875</td>
<td>33.833</td>
<td>332.35</td>
<td>184.433</td>
<td>125.85</td>
<td>87.76</td>
<td>726.40</td>
<td>342.31</td>
<td>2.123</td>
</tr>
<tr>
<td>±0.21*</td>
<td>±0.0381*</td>
<td>±0.459*</td>
<td>±2.756&quot;</td>
<td>±1.576*</td>
<td>±1.308&quot;</td>
<td>±0.468&quot;</td>
<td>±3.5171&quot;</td>
<td>±5.009&quot;</td>
<td>±0.028*</td>
<td></td>
</tr>
<tr>
<td>Ranitidine</td>
<td>2.37</td>
<td>1.0416</td>
<td>34.833</td>
<td>327.783</td>
<td>182.966</td>
<td>124.35</td>
<td>86.933</td>
<td>722.033</td>
<td>343.40</td>
<td>2.1044</td>
</tr>
<tr>
<td>+CNL</td>
<td>±0.19*</td>
<td>±0.65&quot;</td>
<td>±1.452&quot;</td>
<td>±2.4228&quot;</td>
<td>±0.95745&quot;</td>
<td>±1.3788&quot;</td>
<td>±0.42163&quot;</td>
<td>±3.1555&quot;</td>
<td>±4.9962&quot;</td>
<td>±0.027&quot;</td>
</tr>
</tbody>
</table>

Statistical analysis was done using one way ANOVA followed by multiple comparison t-tests. *# significantly different from group I (control) and group III (cerebellar nodular lesion) respectively at p<0.05

134
Histological analysis of combined Alcian blue- PAS stain in stomach and duodenum:

Distribution of the mucopolysaccharide glycoprotein in mucosal layer of control stomach and duodenum tissues is shown Fig 9.1 and Fig 9.6 respectively. Pink and magenta, colour reaction products were glycogen and neutral mucin (highly mucopolysaccharide glycoprotein) respectively.

*C. pepo* extract (400 mg/kg body weight) treatment both the stomach (Fig 9.2) and duodenum (Fig 9.7) showed an increase in glycogen and mucin (mucopolysaccharide glycoprotein) staining reaction products as compared to control stomach (Fig 9.1) and duodenum (Fig 9.6) respectively.

The glycogen and mucin (mucopolysaccharide glycoprotein) reaction products were diminished in the cerebellar nodular lesion (CNL) induced stomach (Fig 9.3) and duodenum (Fig 9.8) tissues as compared to control stomach (Fig 9.1) and duodenum (Fig 9.6). A small amount glycogen and mucin (mucopolysaccharide glycoprotein) reaction product was found in both cerebellar nodular lesion (CNL) induced stomach (Fig 9.3) and duodenum (Fig 9.8) tissues.

Pretreatment of *C. pepo* extract in cerebellar nodular lesion (CNL) induced stomach (Fig 9.4) and duodenum (Fig 9.9) also showed glycogen and mucin (mucopolysaccharide glycoprotein) staining reaction products though less as compared to only *C. pepo* treated stomach (Fig 9.2) and duodenum (Fig 9.7) respectively.

Therefore, pretreatment of ranitidine in cerebellar nodular lesion (CNL) induced stomach (Fig 9.5) and duodenum (Fig 9.10) also showed glycogen and mucin (mucopolysaccharide glycoprotein) staining reaction products though less as compared to only *C. pepo* treated stomach (Fig 9.2) and duodenum (Fig 9.7) respectively.
Histology of stomach and duodenum tissues by combined Alcian blue- PAS stain:

**Stomach: Alcian blue- PAS stain at (×100) magnification**

![Image of stomach histology](image1)

**Duodenum: Alcian blue- PAS stain at (×100) magnification**

![Image of duodenum histology](image2)
Fig 9.1: Shows the distribution of glycogen and mucin (mucopolysaccharide glycoprotein) in mucosal layer of control stomach. Pink and magenta color reaction products were glycogen and neutral mucin (highly mucopolysaccharide glycoprotein) respectively (×100).

Fig 9.2: Shows the *C. pepo* extract treated stomach that increased glycogen and mucin (mucopolysaccharide glycoprotein) as compared with control stomach (×100).

Fig 9.3: Shows that cerebellar nodular lesioned stomach that decreased glycogen and mucin (mucopolysaccharide glycoprotein) as compared with control stomach (×100).

Fig 9.4: Shows the *C. pepo* pretreated in cerebellar nodular lesioned stomach that decreased glycogen and mucin (mucopolysaccharide glycoprotein) though less as compared to only *C. pepo* treated stomach (×100).

Fig 9.5: Shows the ranitidine pretreated in cerebellar nodular lesioned stomach that decreased glycogen and mucin (mucopolysaccharide glycoprotein) though less as compared to only *C. pepo* treated stomach (×100).

Fig 9.6: Shows the distribution of the glycogen and mucin (mucopolysaccharide glycoprotein) in mucosal layer of control duodenum. Pink and magenta color reaction products were glycogen and neutral mucin (highly mucopolysaccharide glycoprotein) respectively (×100).

Fig 9.7: Shows the *C. pepo* extract treated duodenum that increased glycogen and mucin (mucopolysaccharide glycoprotein) as compared with control duodenum (×100).

Fig 9.8: Shows the cerebellar nodular lesioned duodenum that decreased glycogen and mucin (mucopolysaccharide glycoprotein) as compared with control duodenum (×100).

Fig 9.9: Shows the *C. pepo* pretreated in cerebellar nodular lesioned duodenum that decreased glycogen and mucin (mucopolysaccharide glycoprotein) though less as compared to only *C. pepo* treated duodenum (×100).

Fig 9.10: Shows the ranitidine pretreated in cerebellar nodular lesioned duodenum that decreased glycogen and mucin (mucopolysaccharide glycoprotein) though less as compared to only *C. pepo* treated duodenum (×100).

Discussion:

From the results of present experiment, it is evident that treatment with aqueous extract of ripe fruit’s pulp of *C. pepo* (400 mg/kg body weight) decreased acid output, volume of secretion, increased pH of gastric juice, total carbohydrate (e.g. hexose,
hexosamine, fucose and sialic acid) and decreased protein content followed by increased mucus (CHO: P) secretion. But cerebellar nodular lesion (CNL) produced an increase in volume of secretion, excessive gastric acid secretion and decrease in pH, mucus (carbohydrate: protein; CHO: P) content of gastric juice in all rats. The increase in gastric acid secretion (Wolf and Soll, 1988), volume of secretion and decrease of pH, mucous content (CHO: P) of gastric juice may be related to gastric ulcer pathogenesis (Debnath and Guha, 2007; Sarkar et al., 2006; Guha et al., 2002; Sarkar and Guha, 1999; Guha et al., 1995; Guha and Ghosh, 1995; Guha and Maiti, 1996; Maiti and Guha, 1978; Wolf, 1969; Brooks, 1967). The pathophysiology of experimental gastric ulcer formation is not clearly known (Dhikav et al., 2003), so an unified concept for development of gastric lesions by various factors has not yet developed, but it is generally agreed multimechanisms (Goodwin et al., 1986; Konturek et al., 1999) and multifactorial process (Guzel et al., 1998).

The pathogenesis of gastric ulcer disease (GUD) is believed to reflect an imbalance between increased aggressive factors and decreased defensive factors which is partially influenced by defective cerebellum (Sarkar et al., 2006). The involvement of various factors in GUD like an increase in acid secretion (Guha and Maiti, 1990), increase in volume of secretion (Guha and Maiti, 1990), increase in acid-peptic activity (Sonnenberg, 1995), decrease in pH of gastric secretion (Guha and Maiti, 1990), decrease in mucus content (CHO: P) in gastric juice (Debnath and Guha, 2007; Sarkar et al., 2006; Guha et al., 2002; Sarkar and Guha, 1999; Guha et al., 1995; Guha and Ghosh, 1995; Maiti and Guha, 1978; Wolf, 1969; Brooks, 1967), inhibition of mucus secretion, inhibition of bicarbonate (HCO₃⁻) secretion, disruption of mucosal epithelium (Aase, 1989; Allen and Leonard, 1988), inhibition of 5-HT synthesis, reduction of enterochromaffin (EC) cells count and inhibition of prostaglandins (PGs) synthesis in gastro-duodenal mucosa have been suggested (Guha and Ghosh, 1995; Debnath and Guha, 2007; Sarkar et al., 2006; Guha et al., 2002; Sarkar and Guha, 1999; Maiti and Guha, 1978).

The increase in volume of secretion, excessive gastric acid secretion and decrease in pH of gastric secretion, mucus secretion in gastric juice were found after cerebellar nodular lesion (CNL) (Guha et al., 2002; Sarkar and Guha, 1999) by releasing some
endogenous secretagogues such as acetylcholine (ACh) from sub-diaphragmatic cholinergic nerve endings of vagus (Cho and Ogle, 1993; Muallem and Sachs, 1985; Negulescu and Machen, 1988; Soll and Wolling, 1981; Tsunoda et al., 1992), gastrin from antral G-cells (Chew, 1986; Chiba et al., 1988; Chiba et al., 1989; Amagase et al., 1999; Lambersts et al., 1993; Soll et al., 1984; Tsunoda, 1986; Tsunoda et al., 1988) and histamine from mast cells (Sarkar et al., 2006; Bertaccini and Coruzzi, 1989) etc which stimulate the parietal cell to secrete H⁺ ions. These secretagogues are responsible for the increased acid secretion and failure in gastro-protection and repair mechanisms leading to disrupted mucosal barrier by mucosal erosions (Dhikav et al., 2003) and decreased pH of gastric juice (Quigley and Turnberg, 1987).

The gastric mucosa of cardia contains a special type of mucus secreting cells, which has a protective effect by covering the mucosal cells. Mucus serves as a lubricant and also a buffer that traps microorganisms. Mucus mixes with secreted bicarbonate (HCO₃⁻) and helps to keep the pH of the surface of the stomach near neutral levels (Herfindal and Gourley, 1996). The physiological role of mucus is important in mucosal protection and ulcer healing (Allen et al., 1986). Therefore, the gastric mucosa can resist auto-digestion though it is exposed to numerous “insults” like high concentration of HCl, pepsin, reflux of bile, spicy food and microorganisms. So the integrity of gastric mucosa depends on interplay of both defensive and aggressive factors (Diniz et al., 1991). Thus the mucus layer has a direct protective effect by decreasing penetration of noxious agents in deep mucosa through epithelium and maintaining the neutral pH directly above the mucosal cells.

Acetylcholine (ACh), histamine and gastrin interact with muscarinic-3 receptor (M₃) and histamine-2 (H₂) receptor and G-receptor respectively on the parietal (oxyntic) cell and stimulates to HCl secretion by increasing intracellular DAG, IP₃ and Ca^{++} level (Muallem and Sachs, 1985). Cholinergic agents stimulate acid secretion in isolated parietal cell (Batzri and Gardner, 1978; Rosenfeld et al., 1980). The gastric hyper-secretion of hydrochloric acid (HCl) is intimately related to mucosal damage which leads to peptic ulcer disease (PUD) (Wolfe, 1969; Brooks, 1967; Maiti and Guha, 1978).

The increase in volume of secretion, acid secretion, acid-peptic activity and decrease in pH of gastric secretion, mucus secretion in gastric juice observed in the
Present study after cerebellar nodular lesion (CNL) may be due to release some endogenous secretagogues such as acetylcholine, histamine and gastrin etc which stimulated the parietal cell to secrete H⁺ ions. These secretagogues may be responsible for the increased acid secretion and failure in gastro-protection and repair mechanisms leading to disrupted mucosal barrier (as evidenced by mucosal erosions in Alcian blue-PAS staining) and decreased pH of gastric juice in the present experiment.

Treatment with aqueous extract of ripe fruit’s pulp of *C. pepo* showed that 400 mg/kg body weight was the most effective dose (ED) and significantly decreased ulceration and concomitantly increased the mucosal thickness (MT) and alkaline phosphatase (AP) activity which cumulatively showed the defense activity in gastric and duodenal tissues of rats (Sarkar and Guha, 2008). It is also reported that consumption of pumpkin showed a high gastric pH in a rural black African population (Sammon *et al.*, 2003) and the dietetic management of patients undergoing gastric operations is also carried out by supplementation of pumpkin (Loranskaia *et al.*, 1986). The ability of gastric mucosa to resist injury by cerebellar nodular lesion (CNL) is attributed to number of factors that have been referred to collectively as mucosal defense (Guha and Ghosh, 1995). The gastric mucosal erosion induced by cerebellar nodular lesion (CNL) is due to inhibition of this defense mechanisms (Debnath and Guha, 2007).

Many Ayurvedic herbal products (licorice root, chamomile) which increase mucus synthesis accelerate recovery from ulceration (Goso *et al.*, 1996; Beil *et al.*, 1995). It is reported that polyamines have been associated with cell proliferation during ulcer healing (Wang *et al.*, 1990; Brzozowski *et al.*, 1993). A number of studies have demonstrated that polyamines are involved in epidermal growth factors (EGF) mediated gastro-protection, ulcer healing and inhibition of acid secretion (Konturek, 1991; Wojciechowski *et al.*, 1995; Ray *et al.*, 1982; Aihara *et al.*, 1983).

It has been also reported that polyamine-rich food materials of pumpkin as a dietary source of polyamines (Nishimura *et al.*, 2006; Martinez-Tellez *et al.*, 2002). Thus, it may be suggested that pretreatment of *C. pepo* may prevent the gastric mucosal damage (as evidenced by Alcian blue- PAS staining) by increasing mucus synthesis, reducing acid level and peptic activity possibly by increasing epidermal growth factors (EGF) which may be due to the presence of polyamines in the fruit’s pulp of *C. pepo*. 

139
It has been reported that electrolytic lesion in vestibulo-cerebellar area of rat brain produced ulceration and decreased the alkaline phosphatase (AP) enzyme activity present in duodenal tissue of rats (Guha et al., 1995). So, it is clear that normal cerebellum is also involved to prevent gastric ulceration maintaining the activity of alkaline phosphatase (AP) enzyme present in gastric mucosa (Narayan et al., 2004). Therefore, in the preliminary investigations of aqueous extract of ripe fruit’s pulp of C. pepo at a dose of 400 mg/kg body weight exhibited significant protection of gastric and duodenal ulceration by decreasing ulcer index (UI) and concomitantly increasing mucosal thickness (MT) and increasing alkaline phosphatase (AP) enzyme activity present in gastric and duodenal tissues which cumulatively increased mucosal defense activity of gastric and duodenal tissues in all rats against treated with aspirin (Sarkar and Guha, 2008).

Therefore, ripe fruit’s pulp of C. pepo also showed anti-secretory and antiulcer activity by decreasing volume of secretion, acidity and increasing pH, amount of mucus content (CHO: P) (evidenced by biochemical evaluation of mucus content and histological evaluation of Alcian blue-PAS stain) of gastric secretion in cerebellar nodular lesioned (CNL) ulcer model.

In the present investigation, C. pepo caused a significant enhancement of gastric adherent mucus which plays an important role as a defensive factor against mucosal damage, thus confirming the ability of C. pepo to prevent the effects of cerebellar nodular lesion (CNL). These findings indicate that C. pepo pulp extract strengthens the gastric mucosal defense factors in experimental rats.

It is concluded that aqueous extract of ripe fruit’s pulp of Cucurbita pepo Linn. by its anti-secretory and antiulcer property due to presence of anti-secretory and antiulcer components such as vitamin A, C and E, polyamines and their ability to protect gastric and duodenal mucus against cerebellar nodular lesion (CNL) induced excessive acid secretion, excessive acid-peptic activity which lead to peptic ulcer disease (PUD) in rats thus categorising it as an antiulcerogenic herbal drug. However, further studies on different acute and chronic models of ulceration are necessary to rationalise its therapeutic use as an anti-ulcer herbal drug.