SUMMARY

The summary of the work in the thesis entitled "STUDIES ON CONTROL OF THE FORMATION OF HUMIC ACID AND IT'S EFFECT ON KEEPING QUALITY OF SUGAR" is described below in three parts 1, 2 and 3.

In part I, the study on colorants, present in cane juice revealed that it contains natural pigments like chlorophyll, xanthophyll, anthocyanin, and the colorant formed during processing of sugar manufacture.

Spectrophotometer studies of raw, alkaline and acidic cane juice revealed that $\lambda$ max ($\sim$430 to 424 nm) is virtually independent of the medium and corresponds to $\lambda$ max of chlorophyll. However raw cane juice and alkali treated cane juice converse to give $\lambda$ max at 430nm where as the $\lambda$ max of acidic cane juice fall at 425nm. The highest absorbance of cane juice at pH 8 seems to suggest that benzenoid structure of colorant changes to quinonoid structure.

The colorant present in clarified cane juice (Sulphitation process) was isolated by two different methods: - In first method the colorant was isolated from clarified cane juice employing active carbon. The colorant was isolated with the help of azeotropic mixture of water pyridine. A sharp $\lambda$ max 256 nm
in aqueous solution of the colorant seems to be analogous to the \( \lambda \text{ max} \) of humic acid. UV spectra of caramel (\( \lambda \text{ max 280nm} \)) and melanoidin (\( \lambda \text{ max 298 nm} \)) prepared by standard method distinctly differ from the absorption maxima (256 nm) of colorant IR spectra of solid colorant showed a broad peak at 3300 cm\(^{-1}\) of OH group, two peaks at 1590 and 1510 cm\(^{-1}\) of aromatic rings, two peaks at 1250 and 1040 cm\(^{-1}\) of asymmetric and symmetric C-O-C groups, one peak at 1610 cm\(^{-1}\) of quinol group and two peaks at 850 and 790 cm\(^{-1}\) of C-H bending in colorant. The above IR data seem to suggest to this colorant to be closer to humic acid.

In the second method of isolation, the colorant was isolated from deteriorated sugar sample of L-30 (1992-93) by inversion method; the colorant isolated by inversion method is about 4.5 gm, from 100 gm. of cane sugar. A sharp \( \lambda \text{ max 256 nm} \) in aqueous solution and same four peaks were obtained as in case of colorant isolated by employing active carbon, the peaks obtained from the colorant are distinct from the \( \lambda \text{ max} \) of caramel and melanoidin. IR spectra of solid colorant showed a broad peak at 3300 cm\(^{-1}\) of OH group, two peaks at 1590 and 1510 cm\(^{-1}\) of aromatic rings, two peaks at 1250 and 1040 cm\(^{-1}\) of asymmetric and C-O-C groups, one peak at 1610 cm\(^{-1}\) of quinol group and two peaks at 850 and 790 cm\(^{-1}\) of C-H bending in colorant.
The mechanism for the formation of humic acid proposed by W. Flaig seems to be operating in clarified cane juice as well as reducing sugars like hexoses and phenols or polyphenols react to form humic acid.

In acidic medium the absorbance of humic acid remains almost constant whereas in alkaline medium it increases abruptly. This may be due to the fact that in acidic medium quinonic group present in humic acid would have been changed into hydroquinone, which is a benzenoid structure whereas in alkaline medium the hydroquinone of humic acid would change into quinonoid structure.

In order to observe the effect of humic acid on sulphitation reaction, the pH-metric titration of SO₂ solution (Sulfurus acid) was carried out with lime water in the absence and presence of humic acid in cane juice appears to affect both the ionization constant of sulfurous acid. This implies that in presence of humic acid more lime would be necessary for neutralization of SO₂ during clarification reaction.

In part II of the thesis the adsorption of humic acid and Caramel by sugar crystal was studied. In this part, the sugar crystals were developed in the solution of humic acid and caramel and the sucrose solution were examined spectrophotometrically. Adsorption pole of sucrose crystal in the case of caramel was observed, but in case of humic acid no pole of sucrose crystal
was observed, as humic acid is adsorbed uniformly on the sucrose crystal because of colloidal nature and high molecular weight.

The quantity of adsorbed substances in cane sugar crystals is remarkably influenced by pH values of the solution, in which adsorption occurs. The pH value of mother liquor, therefore has an important relation to the quality of cane sugar produced.

In case of caramel, the adsorption of the Caramel increased with pH values of the solution and reaches to its maximum at pH 7, and begins to decrease with further rise in pH.

The adsorbed quantity of humic acid prepared from invert sugar in cane sugar crystals, decreases with pH rise of solution. In cane sugar crystal, the colour of crystals produced, decreases rapidly with pH rise of the mother solutions, and at pH 10, it reaches to almost zero.

Adsorption of humic acid on CaCO₃, CaSO₃, Ca₃(PO₄)₂, Na₂CO₃, Na₂SO₃, Na₃PO₄ and K₂SO₃, K₂CO₃ and K₃PO₄ has been studied. The aim of this part of investigation was to determine the extent of adsorption of the colorant on the above described solids.

In order to observe the absorbability of humic acid, the colorant was allowed to adsorb on the varying amount of the given three (Na, K, Ca) salts
in aqueous and sugar solution separately. It is observed that humic acid is adsorbed comparatively less on Na and K salts. The possible reason for less adsorbability of humic acid could be due to its colloidal nature and higher molecular weight.

The presence of sucrose along with humic acid in solution, greatly influence the adsorption of humic acid on Sodium, Potassium and Calcium salts.

At 25°C the general decrease in adsorption of humic acid in presence of above-mentioned substance could be due to the adsorption of sucrose on Sodium, Potassium and Calcium salts. The Sucrose decreases the available surface of the salts for humic acid adsorption and there by the adsorption of humic acid decreases.

The adsorbing capacity of Na₂SO₃, Na₂CO₃, Na₃PO₄, K₂SO₃, K₂CO₃, K₃PO₄, CaSO₃, CaCO₃, Ca₃(PO₄)₂ for the adsorption of humic acid was also evaluated. The data seem to suggest that Ca₃(PO₄)₂ possesses highest adsorbing capacity among the three set of given salts. It is well known that the surface of solids possesses unsaturated valencies crystal imperfection like dislocation, point defects and grain boundaries. These act as adsorbing centers for colorants. It is expected that the concentration of these surface imperfection in grain boundaries and ionic defects are comparatively higher in
Ca$_3$(PO$_4$)$_2$ than the other two salts, which render high adsorbing capacity of these solids.

Beer's plot of different concentration of humic acid vs absorbance at 420 nm have been utilized to determine the adsorption of humic acid on Sodium, Potassium and Calcium salts in aqueous and sugar solution (15%) separately.

The effect of pH, temperature and concentration on colorant was also studied. The effect of H$^+$ and OH$^-$ ions create a great deflection on the absorbance and structure of the colorant (i.e. in acidic medium the quinone may change into hydroquinone, which is a benzenoid structure, whereas in alkaline medium, the hydroquinone of humic acid may change into quinonoid structure).

To observe the effect of temperature on H$^+$ and OH$^-$ ions, variation in the temperature is done. The absorbance of the colorant increased with temperature.

On addition of different amount of H$^+$ ions, it is observed that the original absorbance of colorant (Humic acid) solution was 0.50 at 25°C, where as at 55° and 70°C the absorbance was observed to be 0.60. On addition of OH$^-$ ions, the original value showed a very much variation than H$^+$ ion. The
effect of OH\textsuperscript{−} ions increased the absorbance value from 0.29 to 0.32, as the temperature increased from 25°C to 70°C.

In case of other colorants such as caramel, melanoidin (Prepared Standard Solution) the effect of H\textsuperscript{+} was observed in case of caramel the minimum absorbance was 0.18.

In case of melanoidin, the absorbance at the three temperatures viz 25°, 55° & 70°C was observed to be 0.80, 0.82, and 0.90 respectively.

The effect of OH\textsuperscript{−} on caramel increased the value up to 0.32 at 70°C after which, no change in the absorbance was observed.

OH\textsuperscript{−} ions did not show any influence on the absorbance of melanoidin, at any temperature.

The comparative adsorbability of humic acid on Na\textsubscript{2}SO\textsubscript{3}, Na\textsubscript{2}CO\textsubscript{3}, Na\textsubscript{3}PO\textsubscript{4}, K\textsubscript{2}SO\textsubscript{3}, K\textsubscript{2}CO\textsubscript{3}, K\textsubscript{3}PO\textsubscript{4}, CaCO\textsubscript{3}, CaSO\textsubscript{3}, Ca\textsubscript{3}(PO\textsubscript{4})\textsubscript{2} were studied. The data suggest, that the least amount of adsorption of humic acid was observed on the potassium salts. Ca\textsubscript{3}(PO\textsubscript{4})\textsubscript{2} was found to be the best adsorbent among all the three salts tested. This suggests that the concentration of dislocation, grain boundaries and ionic defects etc. are comparatively higher on the surface of Ca\textsubscript{3}(PO\textsubscript{4})\textsubscript{2} than the two other salts i.e. (Sodium and Potassium).
The adsorption data of colorant on three salts (Na, K, & Ca) in presence of sucrose suggests that the presence of these salts along with the colorants decreases the adsorption of colorant. The adsorption of humic acid on different solids, as a function of temperature, revealed comparatively higher adsorption of humic acid at 55° and 70°C, particularly in aqueous solution of salts, which seem to suggest the enlargement of surface area of solids.

In part III, the Spectrophotometric and IR techniques for the maximum removal of humic acid. A new method was adopted to remove the humic acid i.e. by centrifuge method rather than, conventional method (Carbonation method), using the two different types of eluents (i) Azeotropc mixture of pyridine (ii) 0.1 M HCl.

In centrifuge method, the raw cane juice was centrifuged at 3000 RPM, to remove the fats and protein, and then this juice was passed through cationic exchange resin and eluted with specific eluent. In case of the conventional method (Carbonation process) humic acid and calcium together act as a buffer, but when the CO$_2$ is passed it forms the carbonic acid, which lowers the pH of the solution and at this stage when the humic acid is adsorbed by calcium particles, comes again in to the solution in the soluble form and
results in the intense colour to the juice, and this intense colour results the degraded sugar.

Part (III) involves determination of molecular weight of humic acid isolated by centrifugation method, done by osmometry process. Osmotic pressure of colorant solution of different concentration were plotted between concentrations of colorant versus $\pi/c$ is given, an average straight line is drawn in the plot, and the straight line has a negative slope which is indication of abnormal behaviour of the colorant in highly concentrated solution. Extrapolation of the straight line to zero concentration gives the value of $\pi/c$, which is $1.01 \times 10^{-4}$ that is equal to RT/M.