EFFECT OF PRE HARVEST BURNING ON SUGARCANE QUALITY AND ON NUTRIENT REMOVAL

By

T.A. KUSUMAWATHIE

A Research Report
Submitted in Partial Fulfillment of the Requirement of
The Diploma in

FOOD SCIENCE AND TECHNOLOGY (FOR CROP SCIENCE)
Affiliated University Collage
Buttala

(Faculty of Applied Science
Uva campus
Sabaragamuwa University of Sri Lanka)

1996

Approved

.....
Externa l Supervisor
Dr. S.A. Ranjith,
Head of Chemistry Division,
Sugarcane Research Institute,
Udawalawe,
Sri Lanka.

Date ..25th Oct. 96.

.....
Internal supervisor
Miss.D.M.I.L.K. Indica,
Lecture in Chemistry,
Faculty of applied science,
University of Sabaragamuwa,
Sri Lanka.

Date 96:10:25.....

.....
Dr.K.K.D.S. Ranaweera,
Act Head,
Department of Physical science,
University of Sabaragamuwa,
Sri Lanka.

Date 25/10-96.

DEDICATED TO

MY EVER LOVING

PARENTS & TEACHERS.

ACKNOWLEDGEMENTS.

First and fore most I would like to thankful to Dr.K.K.D.S. Ranaweera, Act. Head, Department of physical science, Uva campus(buttala), University of Sabaragamuva, for all the services he rendered to me, through out this research.

I would also like to be grateful to my external supervisor, Dr.S.A.Ranjith, Head,Division of Chemistry.Sugarcane Research Institute,Udawalawe,for his valuable guidance,criticisms and support.

I must also thanks Mrs.R.N.T.K.Ubayasena,Research Officer,Division of Chemistry,SRI,Udawalawe,for her invaluable help and guidance throughout my study..

A very special thanks goes to Mr.Bodhinayeke,Research Officer,Division of Chemistry, SRI, Udawalawe, for his kindful co-operation.

I am thankful to my internal supervisor, miss.D.M.I.L.K.Indika, lecturer in Chemistry, Faculty of Applied science, Uva campus,University of Sabaragamuwa, for her advice and assistance.

I also thanks to Dr.U.Ratnaweera. Acting Director,Sugarcane Research Institute,Udawalawe, for giving me permission to carry out this study in the Sugarcane Research Institute.

A special thank is due to staff of Chemistry Division for their valuable co-operation.

I also wish to thank the staff of SRI for their kindness.

I am greatful to prof.A.Bamunuarachchi Consultant to the Uva Campus, University of Sabaragamuwa, for his contribution.

I would like to be thankful to Mr.W.M.S.Rathnappriya, Mr.J.Wansapala, Miss.I.Balam, Miss.K.M.Somawathie. Lectures in Faculty of Applied Science and all members of staff Faculty of Applied Science, Uva Campus, University of Sabargamuwa, for their useful contribution.

A very special thanks to my friend, who helps me in many ways to make this venture suscessfully.

ABSTRACT

An experiment was conducted at the research farm of Sugarcane Research Institute, Uda Walawe to examine the effect of pre-harvest burning on sugarcane quality and on nutrient removal. Twelve months old cane field of variety co 775 was subjected to three harvesting treatments ; i) green cut, ii) burnt, cut and then left lying on the ground and, iii) burnt, and then left standing. Treatments, were replicated five times and were randomized completely. Sugarcane juice were analyzed for total soluble solids(Brix), sucrose content(POL),reducing sugars, dextran, gums and pH for a period of seven days. Weight of cane samples were also recorded. The cane tops and trash left behind after harvesting of green cane and burnt cane as well were weighed separately in each harvesting treatment and analyzed for nutrient contents(Nitrogen,Phosphorus and Potassium).

Cane weight was declined continuously irrespective of the harvesting treatments. Weight loss in burnt cut cane was always greater than the green cut cane. The decrease in POL and purity with time was much higher in burnt standing cane followed by burnt cut cane compared to green cut cane. The decrease of POL and purity were accelerated after two days of burning or harvesting. Reducing sugars,dextran and gums contents were much higher in burnt cane compared with green cut cane. Among the treatments the burnt standing cane had the highest amounts of reducing sugars, dextran and gums. The concentration of dextran and gums, the best indicators of deterioration in sugarcane, were also accelerated after two days of harvesting in burnt cane.

Pre harvest burning of cane also reduced the amount of dry matter and nutrients returned to the field through cane residues.

CONTENTS

LIST OF TABLES

LIST OF FIGURES.

INTRODUCTION.

Sugarcane (*Saccharum officinarum*) a C_4 species. is an economically important crop widely grown throughout the sub tropical and tropical regions of the world.

Sugarcane has been described as the most efficient of all crops in storing the sun's energy (Ledon and Gonzales, 1950). Sugarcane productivity is an integration of various factors. Among the several factors responsible for increasing sugar yield per unit land, suitable varieties, cultural practices and mill efficiencies are ut most important.

The sugarcane plant mainly consists of water, sucrose, reducing sugar, other organic compounds, inorganic compounds, nitrogen bodies, and fibre. the percentage of these constituents depend on variety of cane, its maturity and climate condition.

Sugarcane was introduced into Sri Lanka as a medical herb and now it is grown in Kanthale, Pelwatte, Higurana, and Sevanagala as an industrial crop, to produce sugar. Present production is only 15% of the annual requirementof Sri Lanka which is nearly 488,000 metric tones. The projected demand for the year 2000 is about 490,000 metric tones (Anon. 1995)

The method of harvesting and time gap between harvesting and milling will affect the quality of cane. Sugarcane growers in Sri Lanka practice green cane as well as burnt cane harvesting. Burn cane harvesting has following advantages (Sloane and Rhodes, 1972).

i It reduces cutting cost and time.

ii It facilitates harvesting specially badly lodged cane and fields full of weeds.

iii It eases the preparation of cane at the factory for milling.

iv It increases the milling efficiency since cane containing less trash.

However, when cane is burnt prior to harvesting, the protective wax coating on the outer surface of the stalks is destroyed to a greater or lesser extent. In addition, the rind of the cane may be cracked by the heat of the fire. This allows to increase of naturally occurring micro-organisms such as *Leuconostoc mesenteroides* resulting rapid deterioration of cane, to produce dextran a sugar polymer (Glyn, 1991). When the cane is crushed, the presence of dextran in the cane juice renders the production of sugar very difficult.

Wood (1973) observed that the decline in recoverable sugar is much rapid in burnt and left standing cane than the cane burnt and cut immediately. Therefore, it is recommended that burnt cane must be cut within 24 hours of the fire as burnt standing cane deteriorate much faster than burnt cut cane.

Further more, the burning of sugarcane field causes.

i Loss of parasites of pests.
ii Environmental pollution.
iii Loss of plant nutrient, specialy nitrogen, and organic matter.

Therefore, it is advisable to encourage sugarcane farmers in Sri Lanka to carry out green cane harvesting and to discontinue burnt cane harvesting.

However, there is little information available on the effect of burning on sugarcane quality and on nutrient removal under Sri Lankan conditions. The principal objectives of the present study therefore, to examine the effect of pre harvest burning on sugarcane quality and on nutrient removal.

CHAPTER - 2.

REVIEW OF LITERATURE.

COMPOSITION OF SUGARCANE.

The sugarcane contains, water, sucrose, reducing sugars, organic compounds, inorganic compounds, nitrogenous bodies, ash and fibre(Table.1).

Sucrose occurs in all parts of the sugarcane plant, but it is most abundant in the stalk, where it is found in the watery vacuoles of storage cells.

The generation of reducing sugars by the invert of sucrose represents an undesirable loss in sugarcane processing and is often indicative of other problems, such as formation of dextran, gums etc. (Greenfield and Geronimos. 1985)

Dextrans are products of microbial infection of damaged cells. Dextrans in sugarcane are formed by a bacteria. *Leuconostoc mesentroides*. Dextrans form rapidly under condition of low pH, low Brix and slightly elevated temperature as are found in deteriorated cane juice. The deterioration of chopped cane is accompanied by a rapid increase in dextran (Chen, 1995).

Sugarcane harvesting.

In many sugarcane regions in Sri Lanka, it has become common practice to burn the cane before cutting it. This practice of controlled cane fire, is carried out in order to reduce the amount of trash to be removed manually from the stalk. The advantage of pre harvest controlled cane fire is therefore, a reduction of the cutting cost and time. Depending upon the conditions, one cutter will cut on average of three to six tons of burnt-cane compared to two to there tons of green cane. Since no trash is left on the surface after burning, managment practises, such as irrigation fertilization etc, become easy.

However, several problems are caused by cane burning.There are evidences of sugar loss in burnt cane, particularly if the cane is not cut immediately after burning. This loss, however, is of less significance in subtropical compared with tropical environment (Wood, 1973).

Further, mulching is not possible in those burnt cane fields. In addtion, about one kg of N per tone of cane is lost through this burning process.

However, green cane harvesting causes asubstantial increase in man and equipment hours and the cost of handling of cane. The mill production rate is also decreased by green cane harvesting. It also comes poor sugar rifinability.

Economical gain in increasing sugar yields in green cane harvesting is mastered by the substantial increase in cost of labour machinary and processing in green cane harvesting compared to burnt cane harvesting.

However, burnt cane deteriorates greater than green cane if the harvested cane left behined more than 24 hours. The presence of dextran caused by this deterioration renders the production of sugar very difficult (Glun,1991).

The pH values of juice from green cut cane decline slowly while pH values of juice from burnt cane shows a very rapid decline. This decline is much in burnt standing cane. This indicates the presence of considerable number of acid producing becteria in burnt cane (Waddel,1952). In addition, the burnt standing cane shows the highest lost in sugar. This may be the result of absorption of water from intact root system of burnt standing cane(Egan 1971,Hisa 1972).

Therefore, it is important to transfer cane to the factory within 24 hours of burning to get the economic advantage of burnt cane harvesting. It is always advisable to carry out green cane harvesting. It gives long term benifits through the accumilation of organic matter in the soil.

Table 1. Composition of sugarcane.

Millable cane.	Cane %
Water	73 - 76
Solids	24 - 27
Soluble solids	10 - 16
Fibre (dry)	11 - 16
Juice constituents	**Soluble solid %**
Sugars	75 - 92
Sucrose	70 - 88
Glucose	2 - 4
Frucrose	2 - 4
Salts	3.0 - 4.5
Inorganic acids	1.5 - 4.5
Organic acids	1.0 - 3.0
Organic acids	1.5 - 5.5
Carboxylic acid	1.1 - 3.0
Amino acids	0.5 - 2.5
Other organic nonsugars	
Protein	0.5 - 0.6
Starch	0.001 - 0.050
Gums	0.30 - 0.60
Waxes, Fats, Phosphatides	0.05 - 0.15
Other	3.0 - 5.0

Source: Chen, 1985

CHAPTER - 3.

MATERIALS AND METHODS.

This experiment was carried out at the research farm. Sugrcane Research Institute, Uda Walawa. A twelve months old field of cultivar Co 775 was selected for this study.

The field was divided in to 15 plots of 10m x 16m and subjected to following harvesting treatments after allocating 5 plots randomly for each treatment.

1) cut green.
2) burnt, cut and then left lying on the ground.
3) burnt, and then left standing.

This gave a field design of randomized complete blocks with 5 replicates.

Cane from the 1st and 2nd treatment were bundled after cutting. Each replicate had 7 bundles consisting of 10 stalks for a bundle. Weights of these bundles were recorded until the 7th day of cutting.

A cane bundle from each replicate of a treatment was selected every day until the 7th day of cutting to determine juice quality.

Bundles in the 3rd treatment were prepared by cutting cane just prior to analysis.

Those cane bundles were crushed in a mini sugarcane crusher (Three roller mill - kirloskar model) and juice was extracted and analyzed for,

a) Total soluble solids (Annexure 1)
b) Sucrose content (Annexure 2)
c) pH (Ph meter - TOA PH IM60s)
d) Reducing sugar content (Annexure 3)
e) Dextran content (Annexure 4)
f) Gum content (Annexure 5)

The sugarcane residues such as trash and tops left after harvesting were carefully collected in each plot for the determination of dry weight, total nitrogen (Annexure 6), total phosphorus (Annexure 7), and total potassium (Annexure 8).

CHAPTER - 4.

RESULT AND DISCUSSION.

There was a continuous decline in cane weight after harvest in both unburnt and burnt cut treatments. Weight loss in burnt cut cane was always greater than the unburnt cane (Fig. 1). Loss in weight could be due to loss of water from cane by evaporation. Evaporation rate is highly dependent on climatic factors, such as air temperature, relative humidity, intensity of sunlight, wind velocity etc. Due to the high temperature/heat during burning, the protective wax coating on the outer surface of the stalks is destroyed to a certain extent, and the cane stalks may be cracked and tissues get exposed in burnt cane (Anon 1991). The evaporation of water from burnt cane is therefore, higher than the unburnt cane under the same climatic conditions. It results greater loss in weight in burnt cane.

Changes in percentage total soluble solids in Juice (Brix) with the time is not significant in all treatments(Fig.2). Percentage total soluble in juice is always greater in unburnt cane then burnt cane(Fig.2).

A significant decrease of percentage sugar in the cane was noticed in burnt cane, two days after harvesting compared to unburnt cane (Fig.3). This may be due to the conversion of sucrose. Burnt standing cane showed a greater reduction in percentage sugar in cane with the time. Higher enzymic activitiesdue to high temperature during burning may increase the conversion of sucrose to other products. Such as reducing sugars, This can be major reason for rapid decline in sugar content in burnt cane in addition to the exposure of tissues to decomposing bacteria. In burnt standing cane enzymic activities of the root zone may have resulted in further loss of sugar.

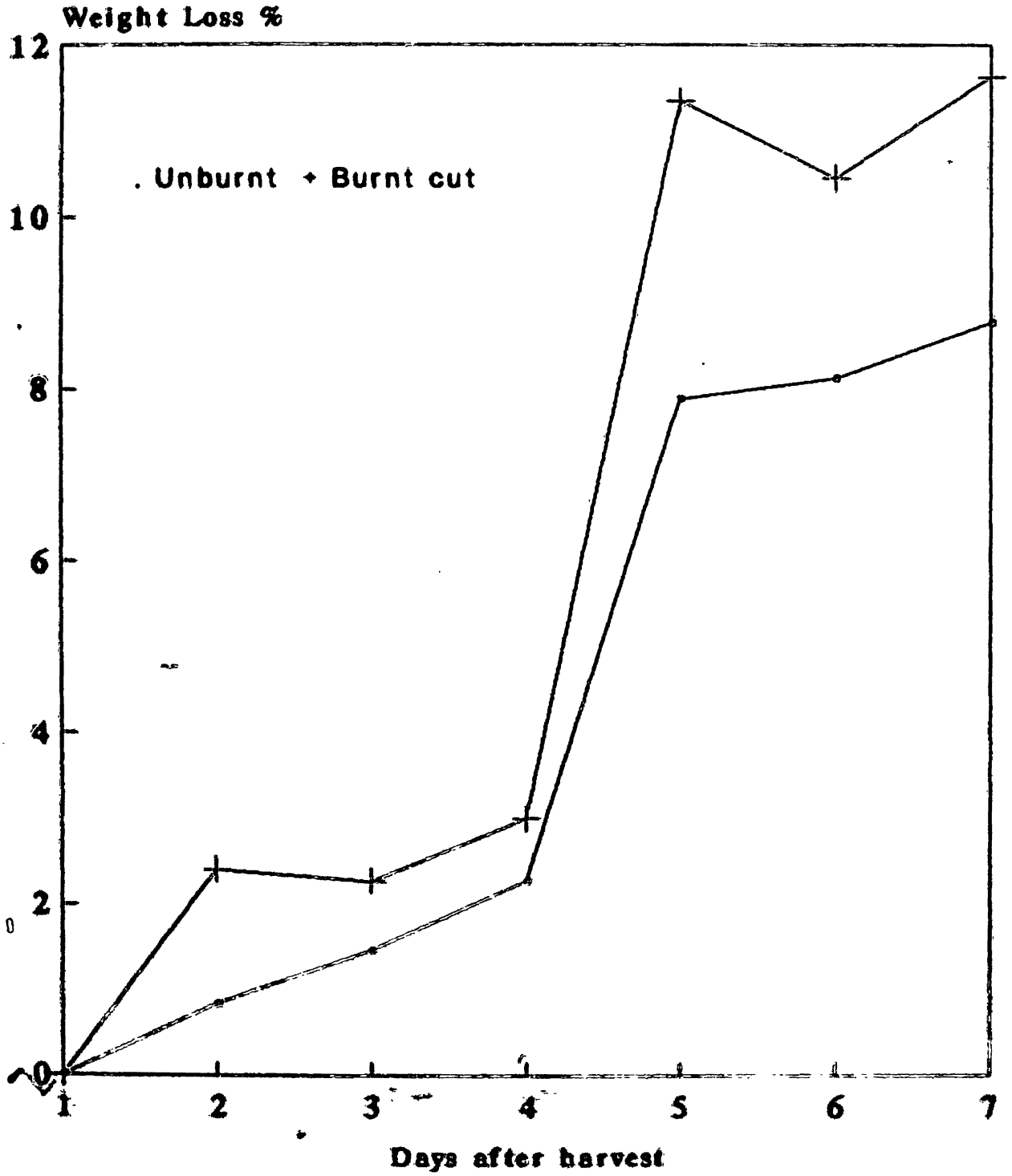

Fig. 1: Percentage weight loss with time

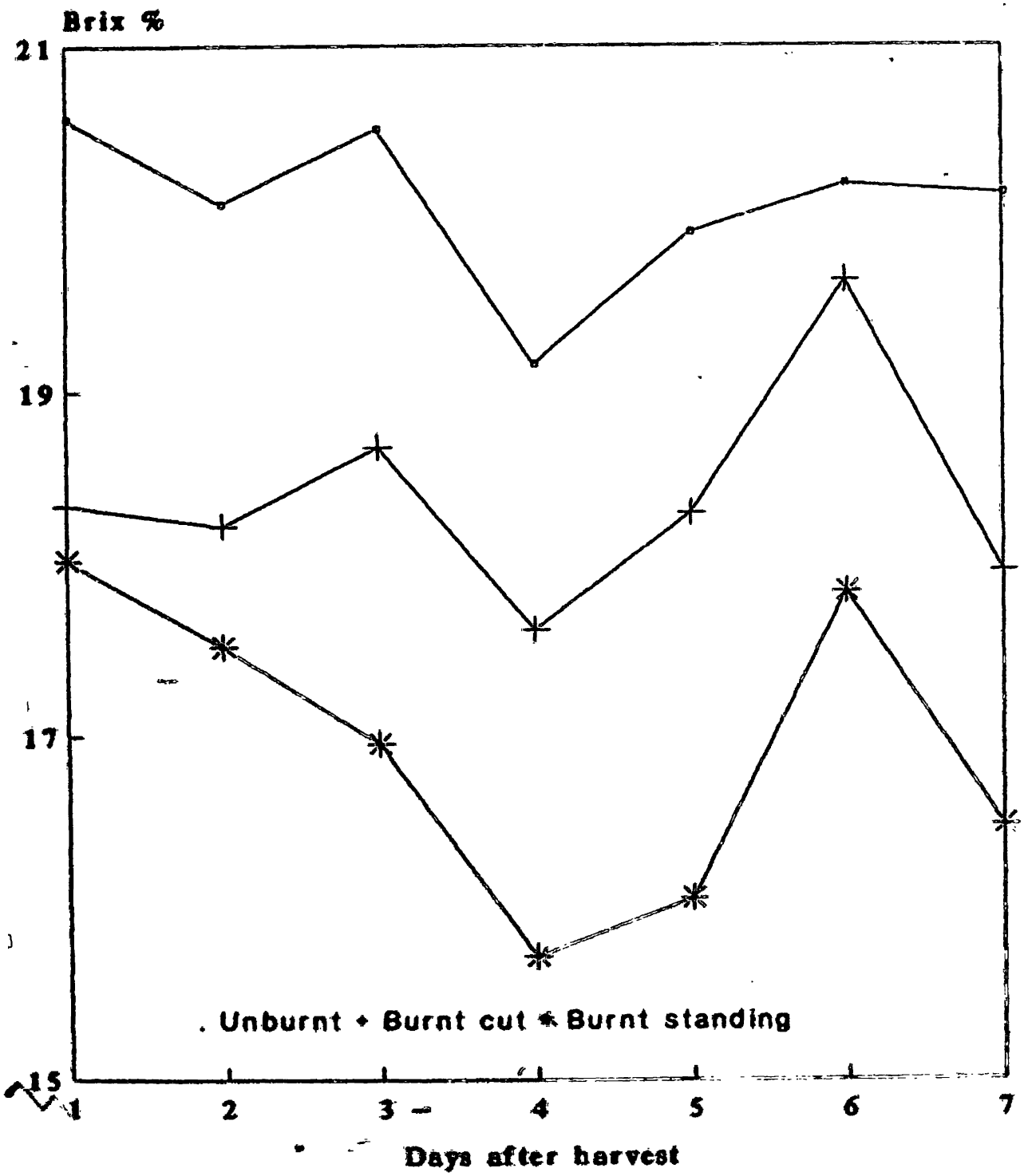

Fig.2:Changes in Brix% with time.

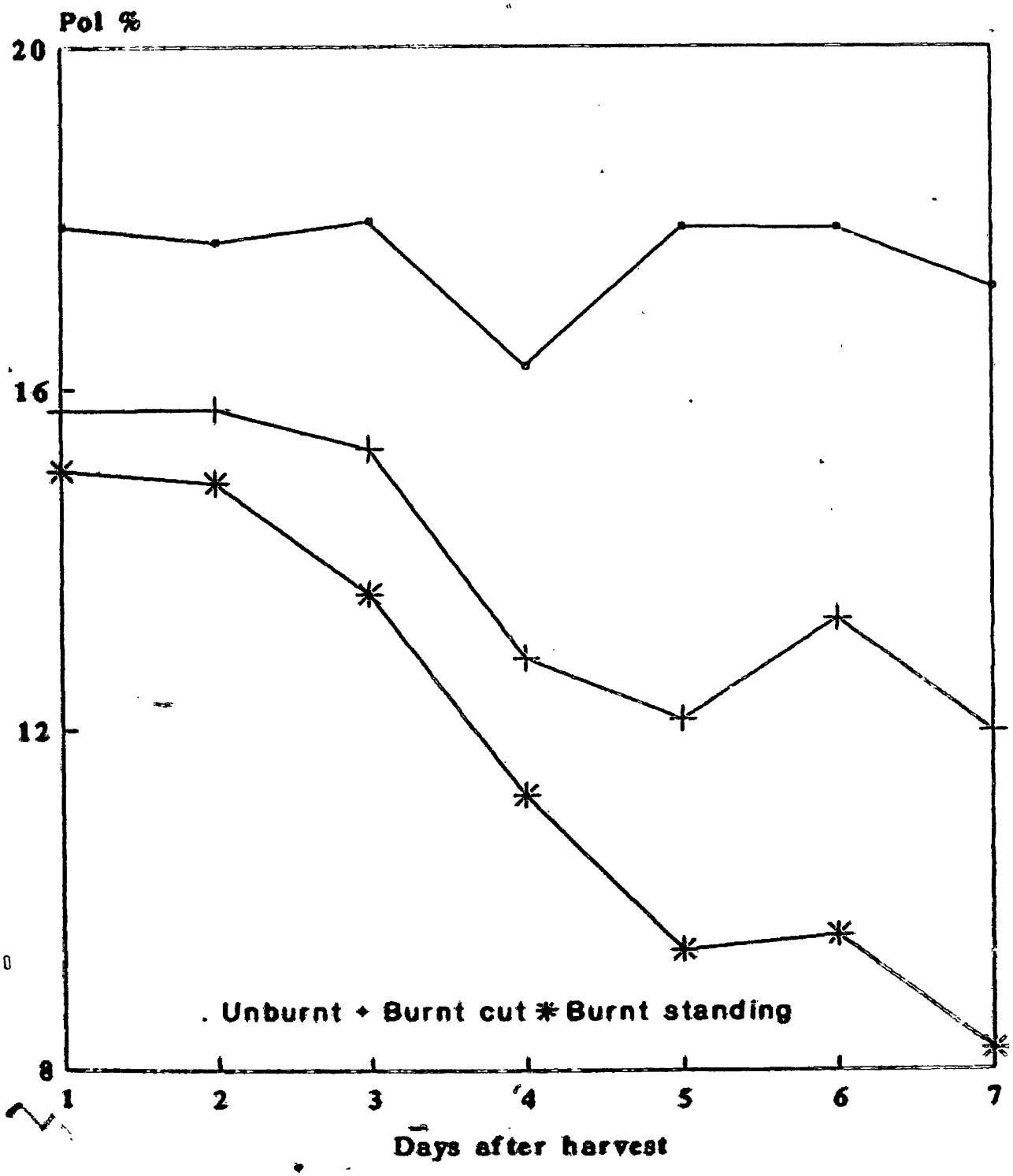

Fig.3:- Changes in Pol% with time.

This loss of sucrose in juice causes lower purity in burnt cane than in unburnt cane (Fig.4). Burnt standing cane showed the highest decline in purity with time.

Rapid increase in reducing sugar content of burnt cane (Fig. 5). may be the main reason for rapid decrease in sucrose content. Increased inversion of sucrose to reducing sugars at a higher rate in burnt cane may be due to increased enzymic activity. In addition, the decrease in pH in juice from burnt cane is the also rapid (Fig.8) and this also accelerates the acid hydrolysis of sucrose in burnt cane.

Rapid decrease in pH in burnt cane juice may be due to the presence of considerable numbers of acid forming bacteria and the possible formation of acetic acid and latic acid(Wood,1972).

Dextran content(Fig.6) and Gum content (Fig.7) in burnt cane increase rapidly with the time compared to that of unburnt cane. This is a result of decomposition of juice by *leuconostoc* bacteria entering the cane stalks through stem ruptures caused by the fire(Wood,1972). Low pH in burnt cane also farves rapid formation of dextran caures many problems in sugar processing including low recovery, increased viscosity with filtration difficulties and crystal destrotion all of which result in loss of final sugar yield.

If was observed that the C.C.S. of green cane after harvesting was always higher than that of burnt cane(Fig.9). After seven days of harvesting, only 1% drop in C.C.S. was observed in green cut cane. Where as in burnt cut cane, the drop in C.C.S. was 4% for the same period. The rate of decrease in C.C.S. was highest in burnt standing cane(Fig.9).

Even four days after harvesting. the C.C.S. of green cut cane was higher than that of burnt cut cane. This is because the deterioration of cane quality starts since the burning in the case of burnt cane where as the deterioration in green cut cane commences just after harvesting .

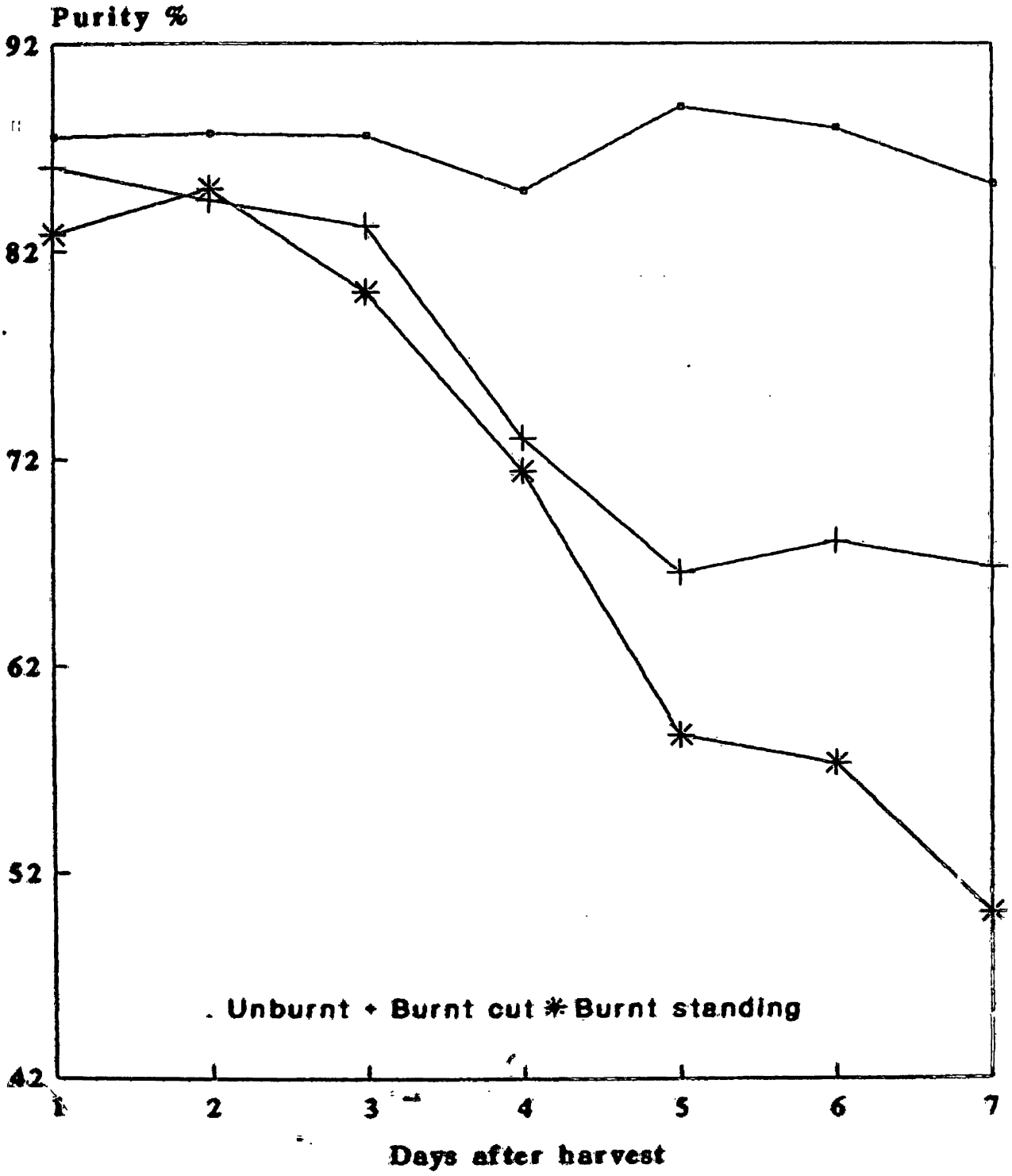

Fig.4:-Changes in Purity% with time.

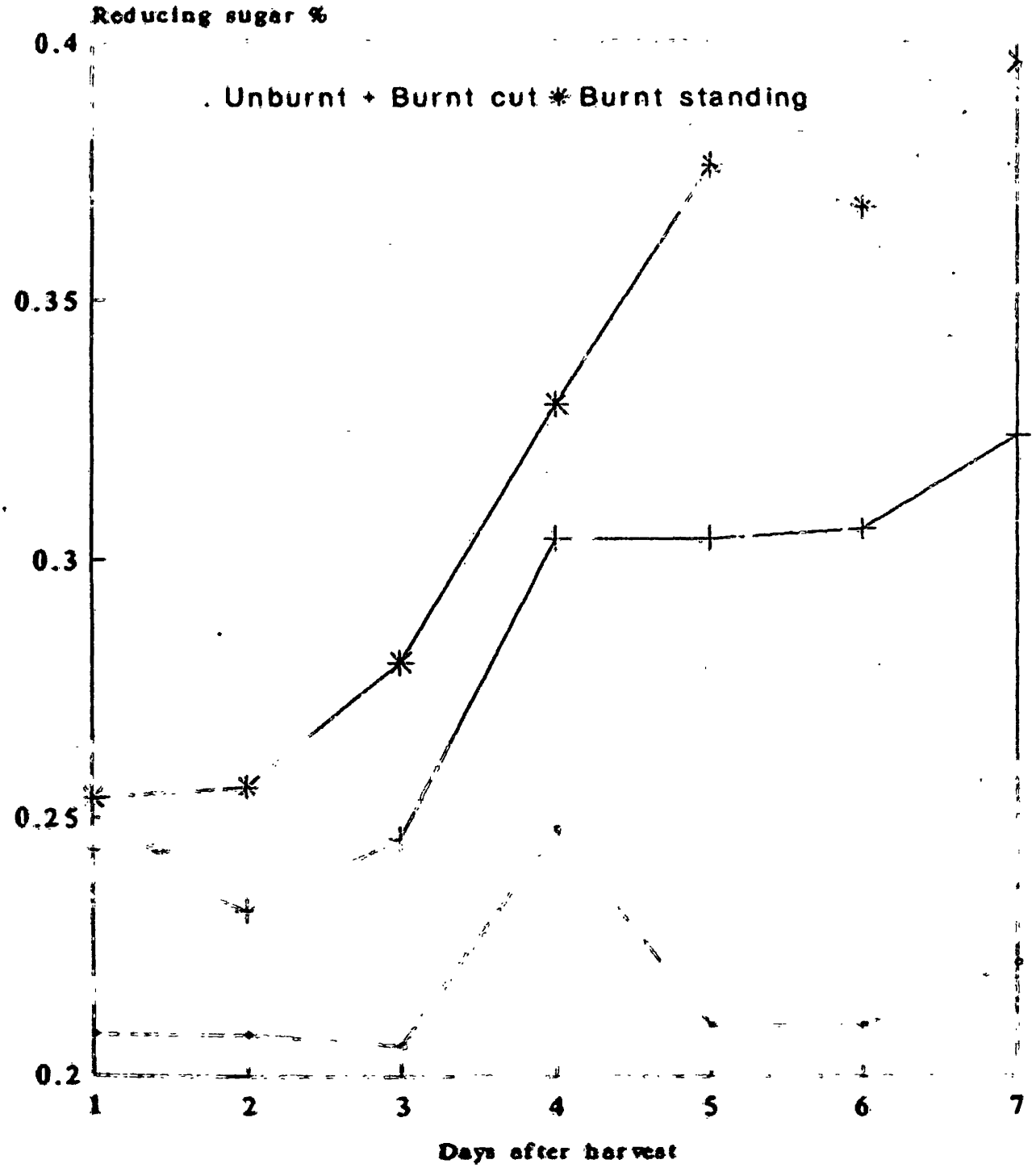

Fig.5:- Changes in reducing sugar% with time.

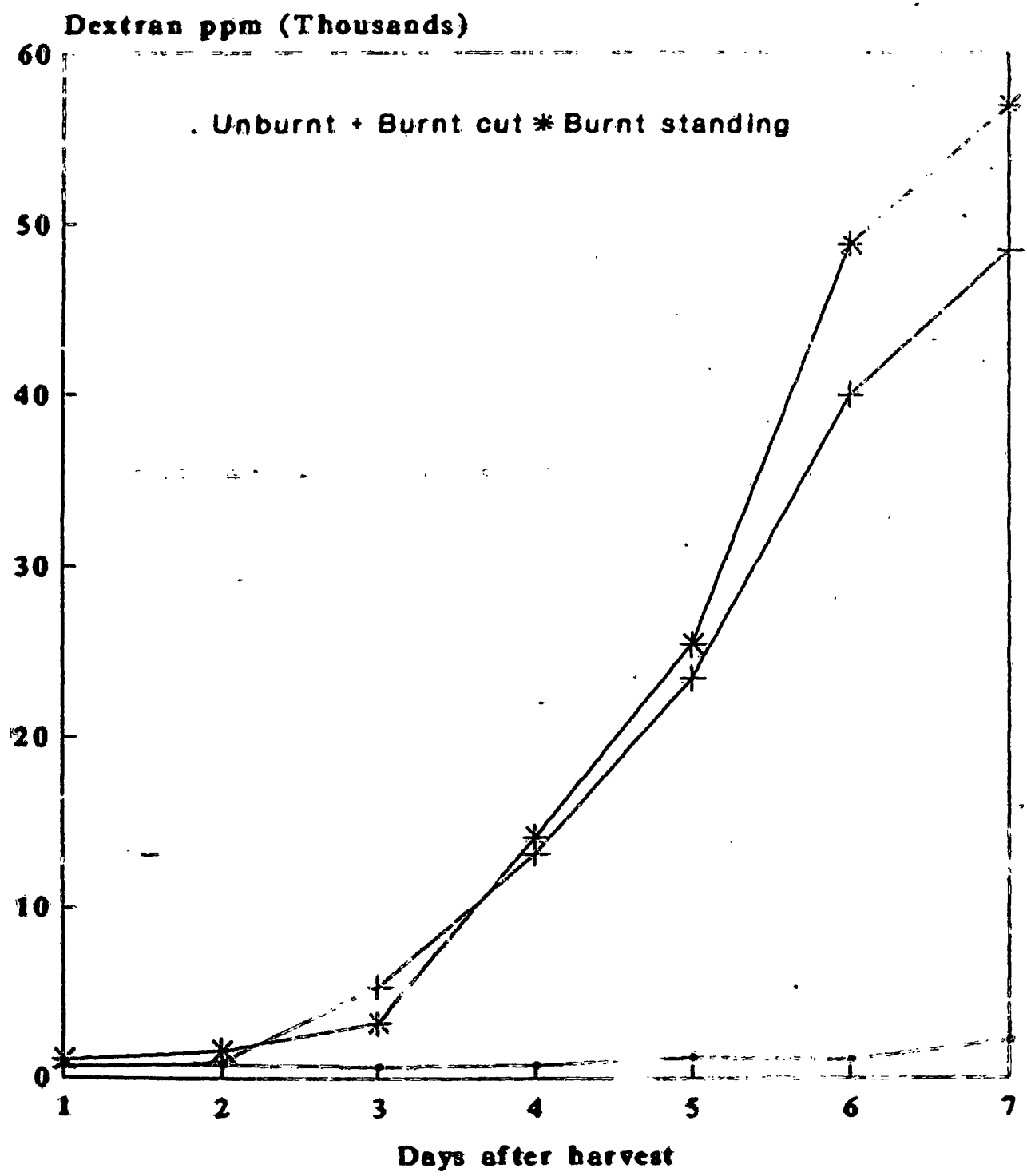

Fig.6:- Changes in dextran content with time.

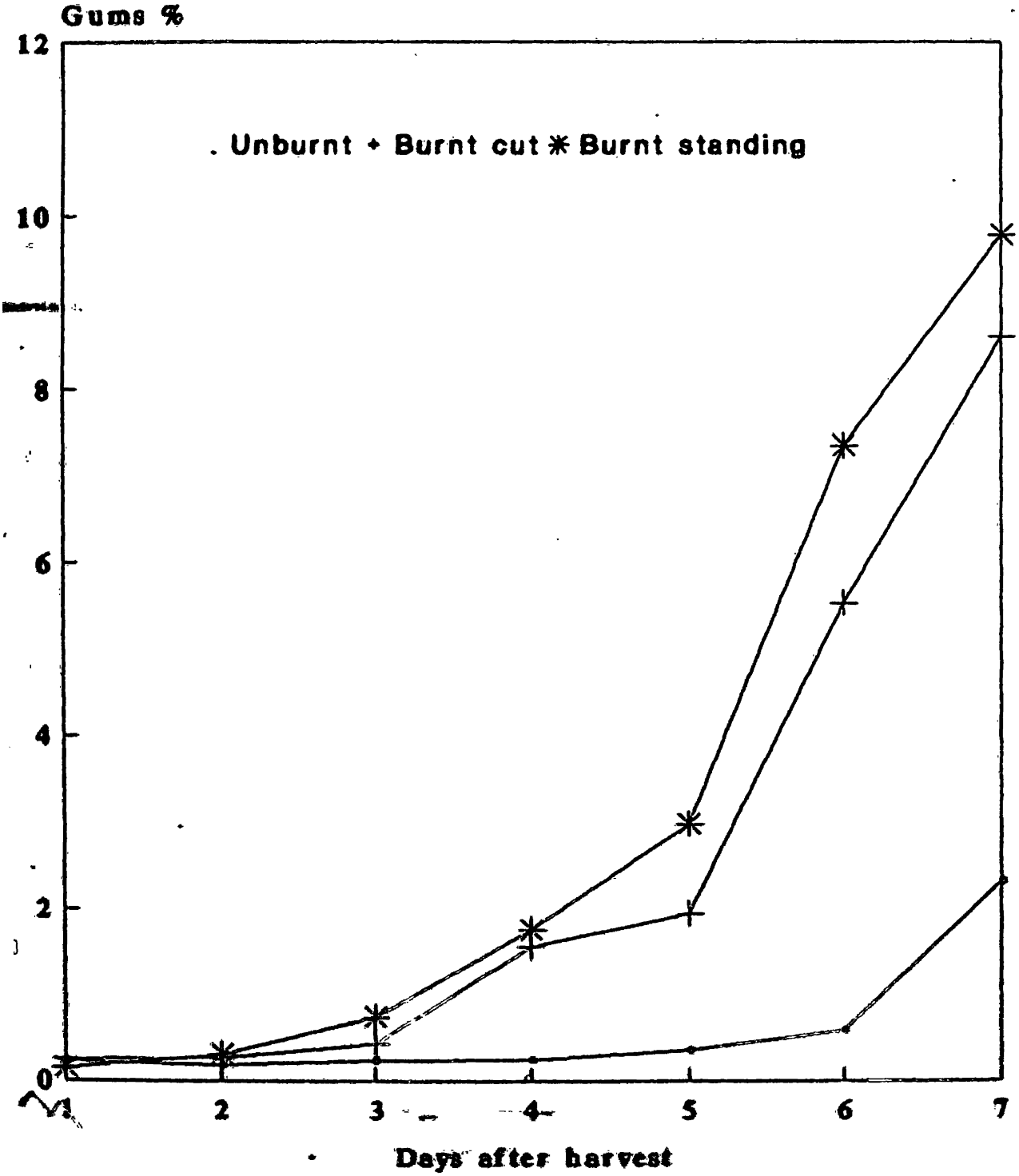

Fig. 7:- Changes in Gum% with time

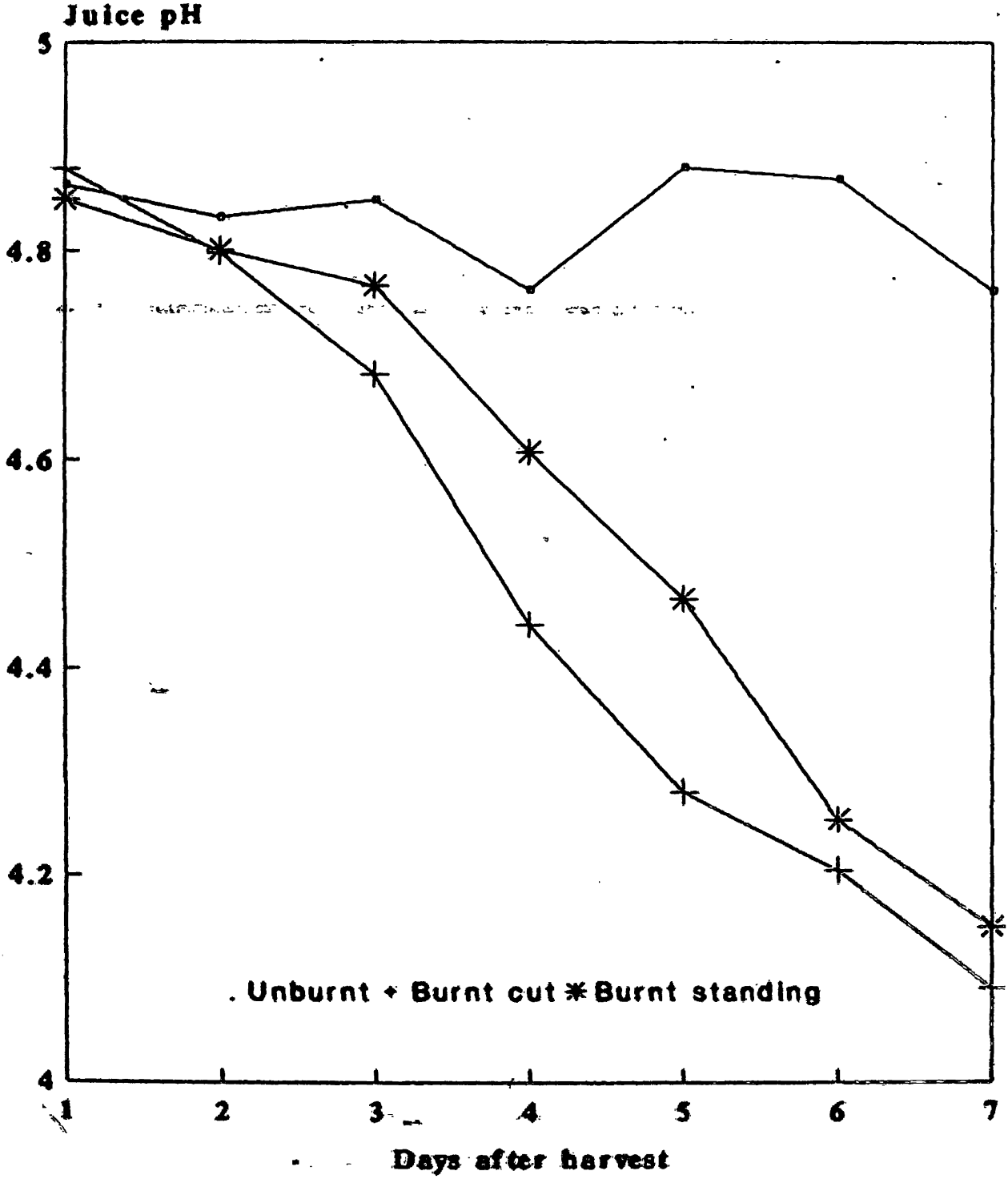

Fig.8:- Changes in pH with time.

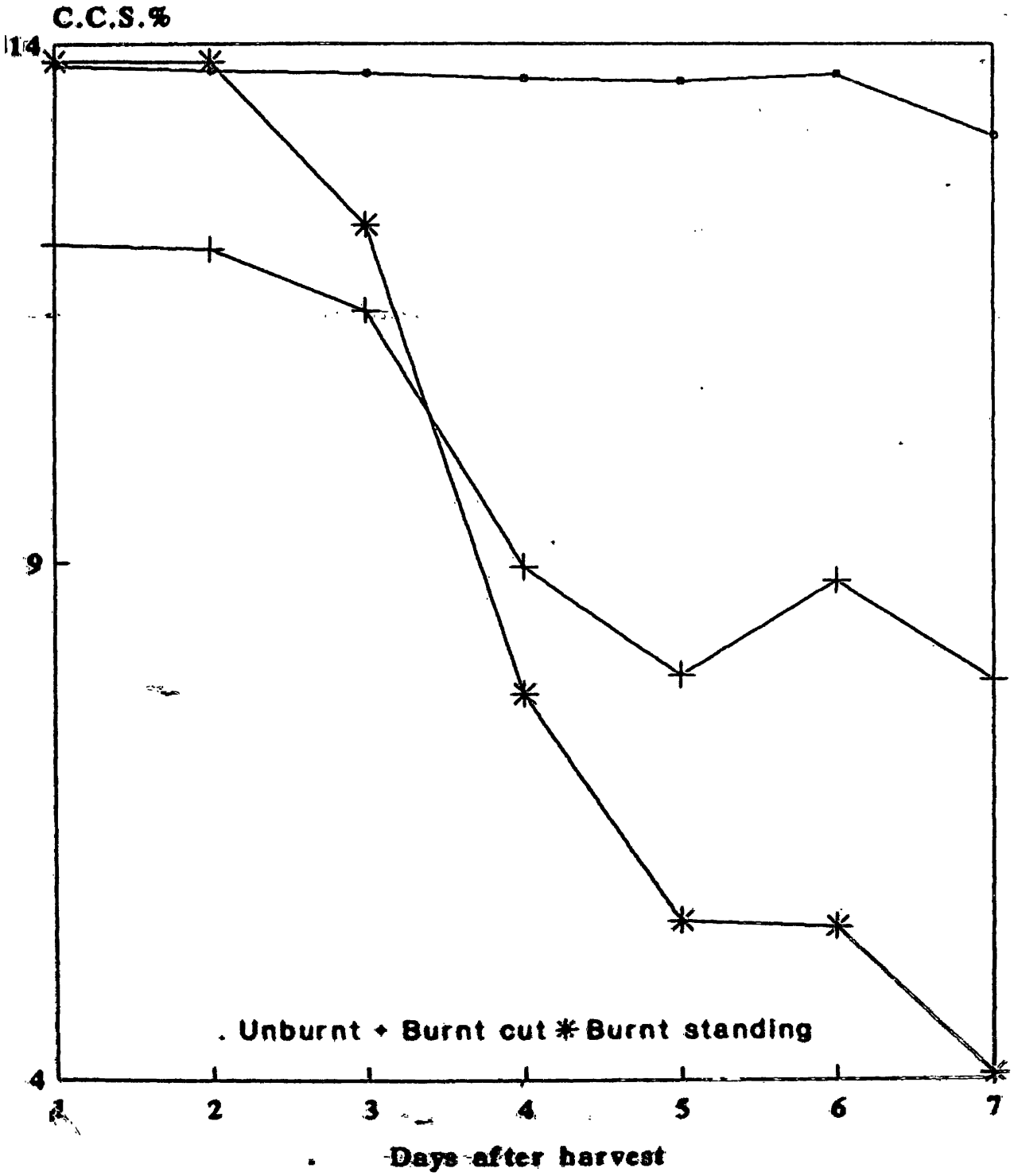

Fig.9:-Changes in CCS with time.

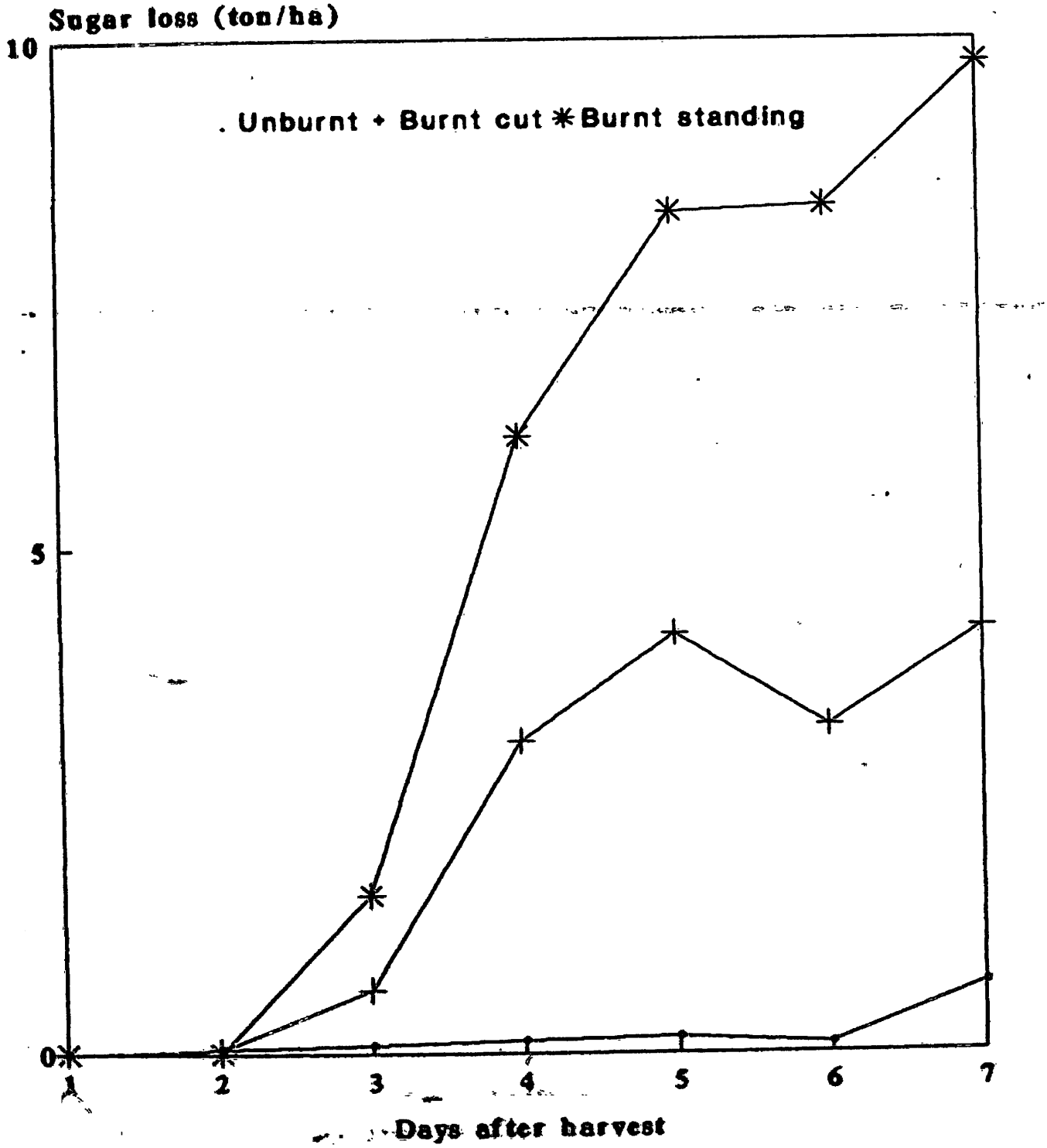

Fig.10: Sugar loss with time.

Therefore, the sugar loss per hectare in green cut cane was not significant during the first 3 days of sampling compared to burnt cane(Table 3).

Therefore, it is always better to carry out green cane harvesting to reduce the sugar loss.

If burning in the only possible meaning of harvesting the cane should be cut immediately after the harvest, because burnt standing cane deteriorates much faster. Hence, it is always advisable to burnt an extent which is harvestable within the same day.

Burning operation at harvesting causes the loss of 34.33 tons of trash per hectare on dry weight losses due to the combustion. this is a sabstational loss of organic matter.

Table 2 Nitrogen, Phosphorus and potassium contents of trash/tops.

Treatments	Nitrogen %	Phosphorus %	potassium %
Unburnt	0.34	0.05	1.12
Burnt	0.29	0.08	1.52

(each value is means of two plots).

Eventhough the differences in N P K content of trash /top of unburnt and burnt cane were not significant (Table 2), about 116.7kg of N/ha is lost in burnt cane harvesting with the loss of 34.33 tons of trash/top. Some of P present in the trash/top can also be also lost during burning.

The substantial amound of residues accumilated after green cane harvesting provide lot of organic matter to the soil, which conserves soil moisture. and improve phycical cemical and biological poperties of soils.

Most of the benificial micro organisms like nitrogen fixing bacteria may get destroyed during the pre harvest burning due to the high tempreture. Burning of cane fileds frior to harvesting also causes many enviromental hazards.

Table 3. Summarized data of the study (each value is mean of five replicates).

	Treatments	DAYS AFTER HARVEST						
		1	2	3	4	5	6	7
WEIGHT LOSS (%)	UN BURNT	0	0.851	1.464	2.282	7.91	8.138	8.77
	BURNT CUT	0	2.406	2.278	3.008	11.35	10.452	11.63
BRIX (%)	UN BURNT	20.581	20.074	20.52	19.16	19.92	20.21	20.14
	BURNT CUT	18.341	18.221	18.68	17.62	18.31	19.64	17.96
	BURNT ST	18.021	17.521	19.96	15.72	16.06	16.84	16.48
POL (%)	UN BURNT	17.902	17.718	17.956	16.264	17.891	17.88	17.181
	BURNT CUT	15.751	15.764	15.302	12.850	12.138	13.31	11.988
	BURNT ST.	15.038	14.898	13.600	11.238	9.418	9.59	8.246
PURITY (%)	UN BURNT	87.524	87.696	87.542	84.882	88.991	87.954	85.232
	BURNT CUT	86.042	84.461	83.236	72.992	66.546	68.042	66.774
	BURNT ST.	82.834	85.031	80.038	71.412	58.696	57.311	50.071
REDUCING SUGAR (%)	UN BURNT	0.208	0.208	0.206	0.248	0.211	0.211	0.222
	BURNT CUT	0.244	0.232	0.246	0.304	0.304	0.306	0.324
	BURNT ST.	0.254	0.256	0.281	0.376	0.376	0.368	0.396
DEXTRAN (PPM)	UN BURNT	707.346	0.772	671.542	800.58	1223.74	1080.66	21 83.
	BURNT CUT	593.998	1003.656	5360.356	13152.08	23406.06	39955.91	48448.42
	BURNT ST.	1041.44	1646.161	3265.236	14176.53	25435.87	48794.78	56966.23

Table 3. summanzed.data of the study (each value is mean of five replicates).

	Treatments	DAYS AFTER HARVEST						
		1	2	3	4	5	6	7
GUMS(%)	UN BURNT	0.221	0.188	0.232	0.246	0.361	0 584	2.318
	BURNT CUT	0.242	0.268	0.428	1.566	1.944	5.536	8.604
	BURNT ST.	0.254	0.304	0.742	1.758	2.986	7 346	9.778
PH	UN BURNT	4.8636	4.833	4.849	4.762	4.881	4.869	4.762
	BURNT CUT	4.8791	4.799	4.682	4.442	4.288	4.205	4.091
	BURNT ST.	4.8504	4.801	4.767	4.608	4.466	4.254	4.151
C.C.S (%)	UN BURNT	13.80	13.75	13.72	13.67	13.64	13.71	13.12
	BURNT CUT	12.07	12.03	11.44	8.97	7.92	8.83	7.87
	BURNT ST.	13.84	13.83	12.26	7.74	5.54	5.48	4.05
SUGAR LOSS (tons/ha)	UN BURNT	0	0.05	0.08	0.13	0.16	0.09	0.68
	BURNT CUT	0	0.04	0.63	3.10	4.15	3.24	4.20
	BURNT ST.	0	0.01	1.58	6.10	8.30	8.36	9.78

Average cane yield under irrigated condition is 100 tons/ha.

CHAPTER - 5.

CONCLUSIONS.

According to the results obtained in this study. following conclusions could be made.

1. Cane weight declined continuously after harvesting irrespective of the methed of harvesting.

2. The decrease in sucrose content and purit with time was much higher in burnt standing cane followed by burnt cut cane than that of green cane.

3. Rate of increase in redusing sugar, dextran and gums contents were much higher in burnt cane compared to green cane. Burnt standing cane recorded the highest amounts of redusing sugar, dextran and gums.

4. Decrease in POL and purity and increase in redusing sugar, dextran and gums were accelerated after two days of harvesting or burning.

5. Burning of cane prior to harvest redused the amount of dry matter and nutrients returned to the field through cane residues.

6. These results indicate that keeping cane in fields more than 2 days after the harvest results in greater economic losses.

7. Steps should be taken to harvest cane immidiately after burning without allowing them to stand.

REFERENCES:

Anon., (199). Annual report, Central Bank of Sri Lnaka.pp.44-47.

Baikow,V.E. (1982). Notes on crop and soil analysis manufacture and refining of row cane sugar. pp.7-11.

Blume,H.(1985). Geography of sugarcane, Tubingengen spring. pp.197-269.

Browne,C.A.and Zerban,F.W. (1941). Physical and chemicals method of sugar analysis. pp.50-56.

Clements,F.H. (1977). Sugarcane crop-logging and crop control principles and practices, University of Hawaii.pp.266-271.

Egan,B.T. (1971). Post harvest deterioration of sugarcane.Bureau of Sugar Experiment Stations,Queensland.

Glyn,J. (1991). Burnt cane gives rise to dextran, a damaging compound. Sugar farming News, SRC, Loutoka, Fiji, bulletin No17.pp.1-10.

Greenfield,P.F. and Geronimos,G. (1985). Measurement of reducing sugars in cane juice. Int.Sug.J.Vol.87,No.1033. pp10-11.

Hisa, Fuu, Yeu. (1972). Effects of burning and storage on cane deterioration. Rept.SES Taiwan. pp.23-56.

Hudson,J.C. (1975). "Green cane harvesting by machine" Paper read to jamaican AST. Sugar producers ,Research unit, Barbados. pp.1-3.

Irvine,J.E. and Legendre, (1973). Deterioration in whole and chopped sugarcane ASSCT,(1973) Louisiana. pp.174-180.

James.C.P.Chen. (1985). Cane sugar hand book,"A Wiley Interscience" New Orleans, Louisiana.pp.6-33,788-798,900-908.

Lisuifong,J.C. "School of industrial technology" University of mauritius. INT SUGAR JNL 1985 vol.84 No.1000. pp.105-108.

Margaret,a.Clarke, et.al (1981), sucrose losses in the harvest of the cane sugar. Crystallizer (TMSI)vol.4 No.1.pp.1-10.

Mettananda,C. (1990). Sugarcane Growing in Sri Lanka, Sugarcane Reseach Institute, Udawalawe.pp.37-51.

Norman,J.King. (1970). Laboratory manual for Queensland sugar mills: Bureau of SES Brisbance.pp.8-42.

Schneider.F. (1978). Sugar analysis ICUMSA methods Pura, Switzerland.pp.1-18.

Sloane,G.E. and Rhodes,L.J. (1972). A comparison of the sugar processing of burnt and unburned sugarcane.Pro.HPRES vol.58,No.14. pp.173-180.

The colonical sugar refining company limited latter to mills and organizations. 14-6-1968.

Williams,T.R. Metalfe,T.R. (1969). "Pest of sugarcane" New York.

Wood,R.A. and Toit,J.L. (1972.) " Deterioration looses in whole stalk sugarcane" Pro.SASTA June 1972. pp.1-7.

Wood,R.A. (1973). "Deterioration losses: Burnt cut,Burnt standing cane. Pro.SASTA June 1973. pp.1-4.

Wood,R.A. (1973). Varietal differences in rate of deterioration whole stalk sugarcane. Pro.SASTA June. pp.1-7.

ANNEXURE

Annexure 1.

Determination of total soluble solids(Brix) in cane juice.

Brix.

The Brix of a solution is the concentration (in g solute per 100g solution) of a solution of pure sucrose in water, having the same density as the solution at the same temperature. If refractive index be adopted as an alternative basis of comparison the value derived should be termed Refractometer Brix.

Procedure

Measure the Brix value of the juice sample using digital Refractometer(ATAGO,RS-1P).

Annexure 2.

Determination of sucrose content (POL) in cane juice.

POL

The pol of a solution is the concentration (in g solute per 100g solution) of a solution of pure sucrose in water having the same optical rotation at the same temperature. For solutions containing only pure sucrose in water, pol is a measure of the concentration of sucrose present.

Reagent

Dry basic led acetate.

Procedure

* Add 1g of dry lead acetate to the 100ml of juice sample.
* Allow the mixture to react for few minutes.
* Filter the prepared solution using Whatman No.91 filter paper.
* Measure the optical rotation of the filterate using digital polarimeter(GASCO,DIP-360).

Calculation:

POL percent:

$$\text{POL percent juice} = \frac{\text{POL Reading}}{\text{POL Factor}}$$

Purity

Purity is the percentage of sucrose in the total soluble solids of a juice sample.

$$\text{Purity} = \frac{\text{POL}}{\text{Brix}} \times 100$$

Commercial Cane Sugar

That percentage by weight of a quantity of cane which would be recovered as pure sucrose if milling and refining operations were conducted at the prescribed standard of efficiency.

Commercial Cane Sugar = $0.82\{P-0.4(B-P)\}$

where,

P = POL percent juice

B = BRIX percent juice

Annexure 3.

Determination of Reducing sugar content in cane juice.

Reagent

1. Fehling Solution
   * Mix together Fehling solution A and Fehling solution B.

2. Methylene blue indicator (1g/100ml)
   * Dissolve 1g of methylene blue in distilled water dilute to 100ml.

Standard invert sugar solution(1g/100ml)

* Dissolve 2.375g of sucrose in 12.00ml in distilled water.
* Add 0.90ml of HCl and then gently mix. The solution is allowed to stand at room tempreture for 8 days.
* After 8 days, add distilled water up to 25ml.
* Check for completion of hydrolysis by a saccharimeter reading.
* 7.15ml of 1N NaOH is add to 20ml of that solution with stirring.
* Dissolve 0.4g of benzoic acid in worm water and then add to the solution mix, cool and make up 200ml.

4. Invert sugar solution (0.5g/100ml).
   * Transfer 50ml of stock solution in to 100ml graduated flask and dilute.

Procedure.

* Mix Fehling A and B solutions to the ratio of 1:1 (by volume) to prepare the Fehlings solution.
* Standardize the prepared Fehling solution using standarded invert sugar solution.
* Pour 10ml of Fehling solution in a 200ml titration flask.
* Add 15ml of juice to the flask using a burette.
* Heat the solution until boiling and continue for 2 minutes.
* Add 4 drops of methylene blue indicator and stire.
* Titrate the reaction mixture quickly using juice until the solution changes to purple to colourless.

Calculation:

Undiluted Juice.

Brix of juice = 19.7
Pol of juice = 17.8
Approximate density of juice = 1.08 g/ml
Sucrose per 100ml
(Pol x Density) = 19.0 g
Titration = 20.0ml
Amount of reducing sugar per 100ml
(from Table) = 222mg

$$\text{percent R.S. in sample} = \frac{222 \times 100}{1000 \times 108}$$
$$= 0.21\ \%$$

Annexure 4.

Determination of Dextran content in cane juice.

Estimation of Haze Intensity:

The turbidity of the dextran haze is determined by reading the "turbidity and color" of the test solution against the "color" of a blank solution in a spectrophotometer set at a wavelength of 720 nm.

Reagents

1. Standard dextran solution (1mg/ ml)
   * Dissolve 0.1g of dextran in 100ml of distilled water.

2. Trichloroacetic Acid Reagent (T.C.A.) (10% w/v).
   * Dissolve 10g of trichloroacetic acid in distilled water , and dilute to 100ml.

3. Denatured Absolute Alcohol (D.A.A.)
   * Mix together 2ml methanol and 98ml ethanol.

4. Standard sugar - standardization solution (50% w/v)
   * Dissolve 50g sucrose in distilled water and Dilute to 100 ml.

5. * Kieselguhr (Purified with acid)

6. * Ion Exchage Resin Mixture

Procedure.

* Obtain calibration curve for Dextran using known concentration of dextran solutions.

* Shake 60ml juice with 2g mixed resin for 10 minutes and filter through 100 mesh gauze.
* Transfer 50ml of juice to a 100ml beaker and add 10ml of Trichloroacetic Acid reagent (10% w/v).
* Add one heaped teaspoonful of kieselguhr to the mixture and stire.
* Filter the mixture (Whatman No.44 filter paper) and transfer 12.5ml of filtrate to a clean, dry 25ml volumetric flask.
* Add Denaturated Absolute alcohol dropwise using a burette and make up to 25ml mark.
* Mix the contents of the flask by inverting gently three times, and allow to mix for 20 minutes.
* Measure finally "absorbance" of the test solution using spectrophotometer at 720nm.

Calculation:

$$\text{Dextran (on solids) (ppm)} = \frac{\text{dextran/25ml (mg)} \times y \times 20}{\text{con (g/ml)} \times \text{aliquot}}$$

where,
y = initial volume of raw cane juice(ml).

Annexure 5.

Determination of Gum content in cane juice.

Reagents

Standard glucose solution.

* Dissolve 0.1g anhydrous glucose(AR grade) in distilled water and dilute to 100ml.

Phenol solution (5%)

* Dissolve 5g phenol(AR grade) in distilled water and dilute to 100ml.

Denatured Adsolute Alcohol solution (D.A.A.)

* Mix together 2ml methanol and 98ml ethanol.

Aqueous Alcohol solution (80% alcohol)

* Dilute 80 parts by volume of Denaturated Absolute Alcohol to 20 parts distilled water.

Procedure

Obtain a calibration curve for standard glucose solution created with phenol and Con. H_2SO_4.
Filter 40ml of juice (325 mesh screen) in to a clean dry centrifuge tube and centrifuged for 10 minutes.
Decant supernatant liquid and read the refractive index (Brix) using refractometer.
Pipette 10ml of centrifuged solution in to a clean 40ml centrifuge tube and add 30ml Denaturated Absolute Alcohol.
Shake the solutions and allow for 35 minutes to form a floc.
Centrifuge for 10 minutes again and decant the supernatant liquid carefully.
Resuspended the precipitate in 40ml of Aqueous Alcohol solution (80% denatured alcohol) and stire.
Shake the solution and allow it for 15 minutes to stand.
Centrifuge the solution for 10 minutes again and decant.
Dissolve the precipitate in 30ml of distilled water in a centrifuge tube and transfer to a 100ml volumetric flask, and dilute to 100ml.

Pipette out 1ml of this solution out in to a test tube and add 1ml of 5% phenol solution with swirling.
Add the 5ml of Con.H_2SO_4 acid and swirl.
Allow the mixture to stand for 10 minutes followed by stirring and cooling to room temperature using a running water bath for further 20 minute.
Measure the absorbance of test solution using spectrophotometer at 485nm wave length.

Calculation:

$$\text{Gums, (\% total soluble solids)} = \frac{\text{glucose(mg) /100}}{\text{concentration(g/ml)x100}}$$

Annexure 6.

Determination of total nitrogen in plant tissue.

Reagents

Salt mixture.

* Mix 250g K_2SO_4 or Na_2SO_4 with 50g $CuSO_4.5H_2O$ and metallic selenium (50:10:1 ratio).

Boric Acid.

* Dissolve 40g H_3BO_3 in 1 liter of distilled water

Mixed indicator .

* Dissolve 0.3g of bromocresol green and 0.2g methyl red in 400 ml of 90% ethanol.

Sodium hydroxide (40%).

* Dissolve 400g of technical grade NaOH in a beaker containing 600ml of distilled water.

5. Standard hydrochloric Acid(0.1N).

* 0.1 N dilute 9ml of concentrated HCl to 1 liter with distilled water. Standardize this approximate 0.1N HCl solution with 0.1N Na_2CO_3 solution.

Procedure.

Place 200mg of dried and ground sample in a micro-kjeldal flask.

Add equal weight of salt mixture and 3ml of con. H_2SO_4 acid. Then digest the sample.

When the sample become clear and cool, add 10ml of distilled water and allow to cool again.

Transfer the digested sample in to the micro-kjeldal distillation apparatus.

Add 10ml of NaOH with quick delivery pipette.

Place 10ml of boric acid and 3 drops of mixed indicator in to 125ml Erlenmeyer flask.

Place this flask under the condenser of distillation apparatus.

Distill the sample for 7 minutes and collect the distillation to the above flask.

Titrate the mixture using standard HCl.

Calculation.

$$\% \text{ nitrogen in sample} = \frac{(a - b) \times N_{HCl} \times 14 \times 100}{w \times 1000}$$

where,

a = sample titer (ml)

b = blank titer (ml)

N_{HCl} = normality of HCl

w = sample weight (g)

Annexure 7.

Determination of total phosphorus in plant tissue.

Reagents

Acid mixture.

* Mix together 70% concentrated HNO_3 and 30% $HClO_4$ (60% - 72%).

Standard phosphorus solution, 50ppm.

* dissolve 0.220g monobasic potassium phosphate (KH_2PO_4) in distilled water and dilute to 1L.

Hydrochloric Acid 1N.

* Add 167ml concentrated HCl to distilled water mix, cool, and bring to 2L.

Nitric Acid 1N.

* Add 125ml of concentrated HNO_3 to 1500ml of distilled water mix, cool, and bring to 2L.

Nitric Acid 2N.

* Add 250ml of concentrated HNO_3 to 1500ml of distilled water, mix, cool, and bring to 2L.

Molybdate vanadate solution.

* Dissolve 25g ammonium molybdate $(NH_4)Mo_7O_{24}.4H_2O$ in 500ml of distilled water.
* Dissolve 1.25g ammonium vanadate NH_4VO_3 in 500ml 1 N HNO_3.
* Then mix equal volumes of these two solutions.

Procedure

Wet Ashing of plant sample.

Place 0.2g of plant material in a 250ml digestion tube.
Add 5ml of the acid and keep stand for overnight.
Transfer the tube to the block digester and heat at 180 ^{0}C until the material is digested (2 hour)
Remove the tubes from the block and allow to cool.
Add 5ml of 1N Hcl and bring to 50ml with distilled water.
Filter a portion of the solution through a Whatman No.40 filter.

Color development.

* Transfer 1.00ml aliquot of the wet ash solution in to a 10ml of volumetric flask.
* Dilute 2ml of 2N HNO_3 to 5ml with distilled water.
* Add 1ml of the molybdate - vanadate solution and make upto 10ml with distilled water.
* After shaking allow it to stand for 20 minutes.
* Measure the absorbance of samples using spectrophotometer at 420 nm.

Calculation:

$$\% \text{ P in sample} = \frac{(a - b) \times 50ml \times 100}{10,00000 \times w}$$

where,

a = sample reading in ppm
b = blank reading in ppm
w = sample weight (g)

Annexure 8.

Determination of total potassium content of plant material.

Transfer 1ml aliquot of the wet ash solution in to a 10ml volumetric flask and dilute to 10ml using 1N HCl. Measure the absorbance of sample using Atomic absorption spectrophotometer at 765.5 nm.

Calculation:

$$\% \text{ K in sample} = \frac{(a - b) \times 50\text{ml} \times 10\text{ml} \times 100}{10,00000 \times w}$$

where,

a = sample reading in ppm

b = blank reading in ppm

w = sample weight (g)