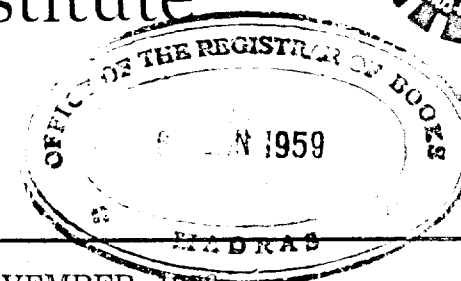


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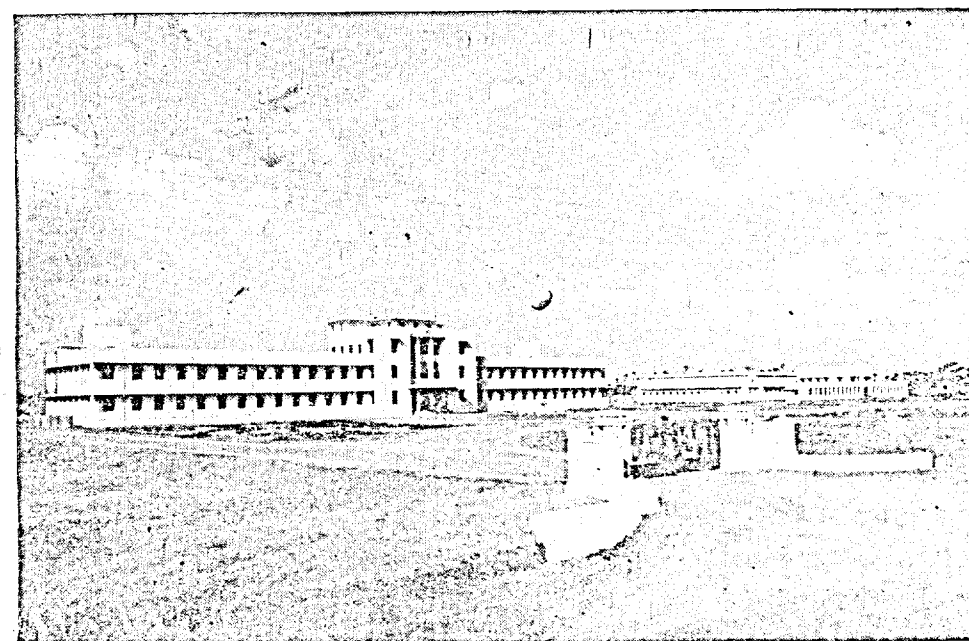
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INDIA

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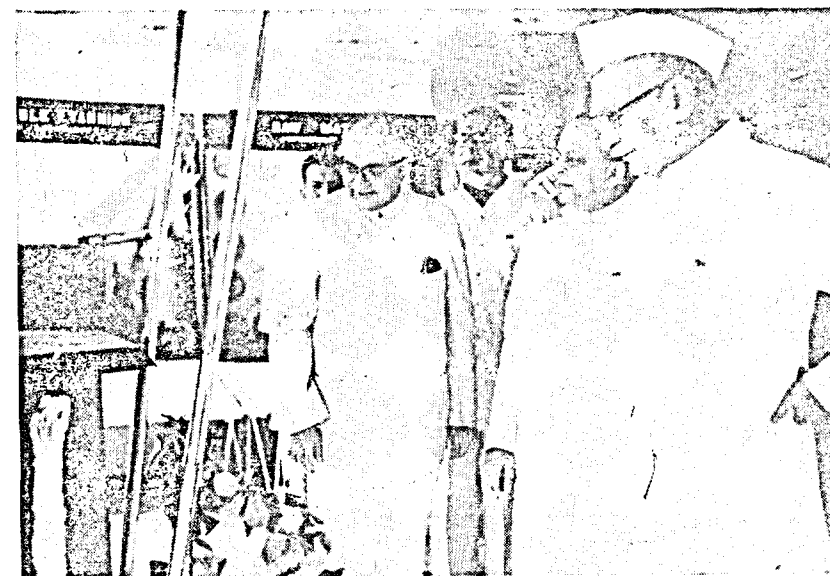
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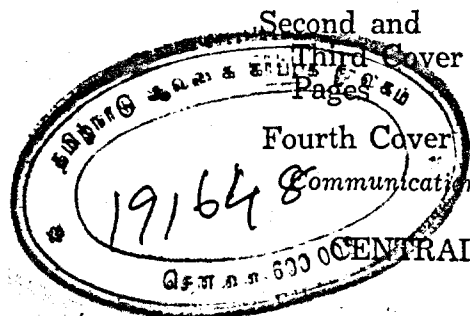
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Dr. Rajendra Prasad, President, Indian Union, at the CLRI Stall, in the India—1958 Exhibition. To his right are seen, Shri N. Kanungo, Minister for Commerce, and Prof. M. S. Thacker, Director-General, Scientific and Industrial Research

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INTERACTION OF COLLAGEN WITH CERTAIN PHENOLS-HEXAMETHYLENE TETRAMINE COMPLEXES

BY

S. RANGANATHAN, S. M. BOSE & Y. NAYUDAMMA

Central Leather Research Institute, Madras

(Received on 5th April, 1958)

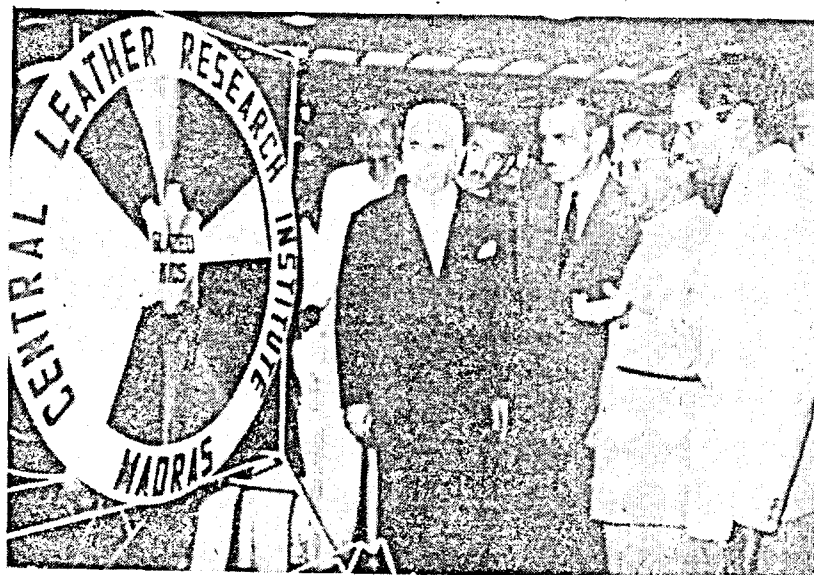
SUMMARY

Complexes of resorcinol, catechol and phenol with hexamethylene tetramine have been prepared and their interaction with collagen studied. The tanning effect of resorcinol-hexamethylene tetramine complex was found to be due to its cleavage to resorcinol, formaldehyde and ammonia and the subsequent linking of resorcinol and formaldehyde with the primary amino groups of collagen involving the formation of Mannich linkages. The reaction was found to take place at almost all pH levels but mostly from 3-8, where polymerisation was least.

In the course of a study of the properties of hexamethylene tetramine, a compound prepared from hexamethylene tetramine and resorcinol was found to possess good tanning properties.¹ The leathers produced had a high shrinkage temperature, suggesting the formation of stable linkages with collagen.

Preliminary experiments showed that during tanning the complex was cleaved to a certain extent to resorcinol, formaldehyde and ammonia and that only resorcinol and formaldehyde were involved in tanning.

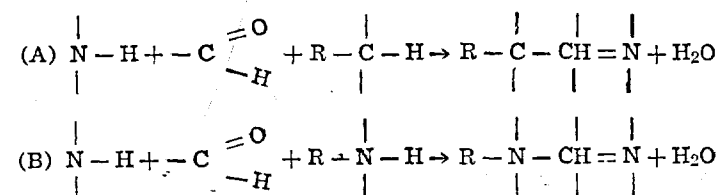
In 1952, Windus² found that a leather having a high shrinkage temperature was obtained by treating pelt with resorcinol and formaldehyde. He postulated that the tanning involved the formation of Mannich linkages. It was considered desirable to make a detailed study of the reaction of resorcinol-hexamethylene tetramine complex with col-



Prof. Humayun Kabir, Vice President, Council of Scientific and Industrial Research and Minister for Scientific Research and Cultural Affairs, at the CLRI Stall, in the India-1958 Exhibition

lagen in order to get an insight into the nature of reaction, that is taking place.

The Mannich reaction³ may be represented in a generalised way as follows :



The reaction A produces a C-Mannich base and B produces a N-Mannich base. The formation of the latter type of linkage, during tanning of proteins was first positively established by Nitchman and Hadorn⁴ and the subsequent exhaustive study made by Fraenkel-Conrat and his co-workers,⁵⁻¹⁰ established beyond doubt the formation of -N-CH₂-N- linkages during tanning of proteins with formaldehyde. In the case of tanning of collagen with formaldehyde, the ε-amino groups of lysine, and the guanidino groups of arginine to a certain extent, can form cross links with the amide or the guanidino groups of the protein. However such a N-Mannich base linkage is easily susceptible to hydrolysis and naturally the formaldehyde bound could be quantitatively hydrolyzed by strong acids.

A C-Mannich base differs from the N-Mannich base in that it is comparatively much more stable and that hydrolysis does not take place easily. Fraenkel-Conrat and his co-workers⁹ showed that large amounts of formaldehyde were irreversibly bound when gramicidin, a protein rich in tryptophane, was treated with formaldehyde and *p*-cresol. It was further shown that indole, threonine, histidine and proline bound formaldehyde in an irreversible way when treated with *p*-cresol. They concluded that a C-Mannich reaction was taking place.

In the case of resorcinol-hexamethylene tetramine complex, it was found that some amount of resorcinol and

formaldehyde were bound irreversibly by collagen during the reaction. A detailed study of the effect of pH and the nature of collagen on the fixation of complex has been made. A quantitative study of the reaction of resorcinol and formaldehyde with collagen at various pH values also has been carried out in order to find out the effect of pH on the course of the reaction.

Other compounds which were tried for tanning action were the hexamethylene tetramine complexes of phenol, catechol and pyrogallol. All these compounds excepting the phenol complex had tanning action in varying degrees. A somewhat detailed study of the reaction by phenol-hexamethylene tetramine complex and catechol-hexamethylene tetramine complex also has been made in this connection. To find out the effect of formaldehyde donor, hexamethylene tetramine was replaced by bis-hydroxymethylurea.

EXPERIMENTAL

Preparation of Complexes: Generally the complexes were prepared by mixing equimolecular amounts of the phenols and hexamethylene tetramine taken as saturated aqueous solutions. In the case of phenol-hexamethylene tetramine complex the phenol and hexamethylene tetramine were taken in the molar ratio 3 : 1.

The yields and melting points of the complexes are given below :—

TABLE I

No.	Complex	m.p.	yield
1.	Resorcinol-Hexamethylene tetramine complex.....	178°C	90%
2.	Phenol-hexamethylene tetramine complex.....	125°C	98%
3.	Catechol-hexamethylene tetramine complex.....	150°C	70%
1.	% N calcd. for C ₆ H ₆ O ₂ (CH ₂) ₆ N ₄	22.4; % N found	22.6.
3.	-Do-	22.4; % N found	22.7
2.	mp. lit. 124° [for 3C ₆ H ₆ O. 1(CH ₂) ₆ N ₄]		

Estimation of complexes. In all the cases the phenols were estimated by the bromide-bromate¹² method. In the case of catechol-hexamethylene tetramine complex the estimated amounts were always found to be lower than the actual amounts. This may be due to incomplete bromination. So in this case a graph was drawn with the actual amounts used against the corresponding volume of standard thiosulphate consumed. For estimation of unknown quantities the estimation was done under identical conditions as employed for drawing the standard graph and by knowing the exact amount of sodium thiosulphate consumed the amount of the complex present was read out from the straight line graph. The accuracy of estimation of phenol and resorcinol from the complexes was tested by taking known amounts of complexes.

TABLE II

No.	Complex	Amount taken	Amount found
1.	Resorcinol-hexamethylene tetramine complex	20.4 mg.	20.9 mg.
2.	Phenol-hexamethylene tetramine complex	20.1 mg.	20.5 mg.

With resorcinol hexamethylene tetramine complex, methods for estimation of formaldehyde and ammonia also were standardized. Ammonia was estimated by heating the complex with *N/70* sulfuric acid for 0.5 hr. and then back titrating the excess acid against standard *N/70* sodium hydroxide solution. Formaldehyde was estimated by the bisulphite method.¹³ The accuracy of the methods was tested by employing known amounts of the complex.

Calculated ammonia that could be obtained from 6.8 mg. of complex : 1.8 mg., found 1.9 mg.

Calculated formaldehyde that could be obtained from 6.8 mg. of complex : 4.9 mg., found 5 mg.

Interaction of resorcinol-hexamethylene tetramine with pelt :

In a solution prepared from 770 mg. of the complex and 770 mg. of sodium chloride in 14 ml. of water was placed 7.7 g. of pelt, the pH adjusted to 6 and the mixture allowed to stand at room temperature for 40 hours with intermittent shaking. The pelt was then cut into small pieces and extracted repeatedly with water. The combined washings and the residual liquor were made up to 2 l, and analysed for ammonia, formaldehyde and resorcinol. The leather was heated on a boiling water bath for 0.5 hour with *N/70* sulfuric acid and the solution analysed for ammonia, formaldehyde and resorcinol.

TABLE III

Compound	Calcd./ 770 mg. complex	Found		Total	Difference
		from washings and residual liquor	from leather		
Ammonia	209.4	191.4	21.9	213.3	nil.
Formaldehyde	554.6	394.8	15.44	410.2	144.4
Resorcinol	338.8	198.1	11.7	209.8	129.0

Reaction of hexamethylene tetramine and hexamethylene tetramine-complex with glycine :

An aqueous solution prepared from hexamethylene tetramine (500 mg.) sodium chloride (500 mg.) and glycine (500 mg.) was adjusted to pH 6.5 and allowed to stand at room temperature for a period of 40 hours with intermittent

shaking. The solution was then analysed for ammonia and formaldehyde. The above experiment was repeated using 500 mg. of resorcinol-hexamethylene tetramine complex for hexamethylene tetramine. The solution was then analysed for resorcinol, formaldehyde and ammonia.

TABLE IV

Compound	hexamethylene tetramine			resorcinol-hexamethylene tetramine complex		
	calcd./500 mg.	found	differ-ence	calcd./500 mg.	found	differ-ence
Ammonia ..	242.8	237.3	5.5	136.0	136.0	—
Formaldehyde ..	642.7	625.0	17.7	360.0	174.4	185.6
Resorcinol ..	—	—	—	220.0	75.6	144.4

Interaction of resorcinol hexamethylene tetramine complex with (a) collagen, (b) deaminised collagen and (c) ultramid 6A and stability, of the complex in aqueous medium:

A solution prepared from 200 mg. of pure complex and 200 mg. of sodium chloride in water (15 ml.) was adjusted to pH 3 and kept at room temperature for 40 hours with intermittent shaking. The above experiment was repeated using the same amounts but at pH values from 4 to 10. The unreacted resorcinol was then estimated in the usual way.

The blank experiment was repeated using 2 g. lots of collagen, deaminised collagen and ultramid 6 A. After keeping for 40 hours the collagen or deaminised collagen or ultramid 6A was repeatedly washed with water, the total volumes made up and resorcinol estimated as usual. The

net amount used up in reaction was plotted against the pH values; the results are presented in Figures I and II.

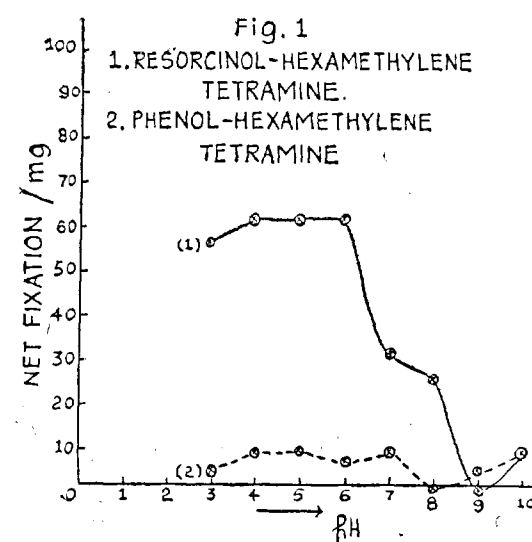
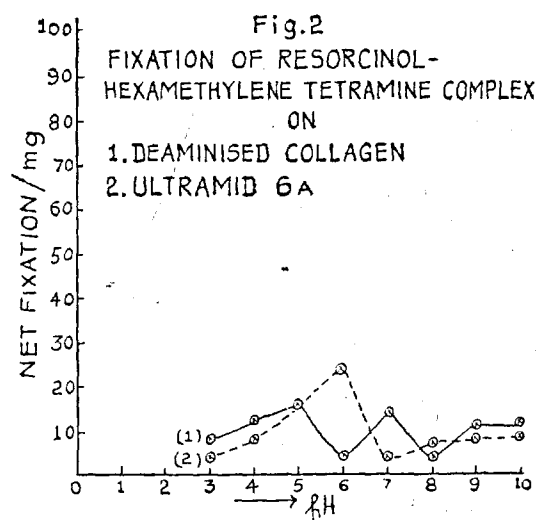


TABLE V

200 mg. Complex; 40 hours; 2 g. collagen or 2 g. deaminised collagen or 2 g. ultramid 6A.

pH	Amount of complex used up/mg.			
	Blank	Collagen	Deaminised collagen	Ultramid 6A
3	7.1	62.85	15.7	11.4
4	2.8	62.85	15.7	11.4
5	2.8	62.85	20.0	20.0
6	2.8	62.85	7.0	27.1
7	2.8	32.9	15.7	7.1
8	7.1	32.9	11.4	14.6
9	15.7	9.3	27.2	24.2
10	15.7	24.3	27.2	24.6



The reaction of resorcinol and formaldehyde with collagen: The stability of resorcinol-formaldehyde mixture at various pH levels:

A solution prepared from 160 mg. of resorcinol, 130 mg. of formaldehyde (as 33% solution) and 320 mg. of sodium chloride, in water (20 ml.) was adjusted to pH 2 and allowed to stand at room temperature for 20 hours with intermittent shaking. The above experiment was repeated using same amounts of reactants but at pH values from 3 to 10. After reaction the unreacted resorcinol was estimated. The blank experiment was repeated using 2 g. lots of collagen from pH 2 to 10. After allowing to stand for 20 hours the solutions were filtered, the collagen washed repeatedly with water, the total washings made up and resorcinol estimated as usual. The net fixation was plotted against pH values and the results are presented in Figure III.

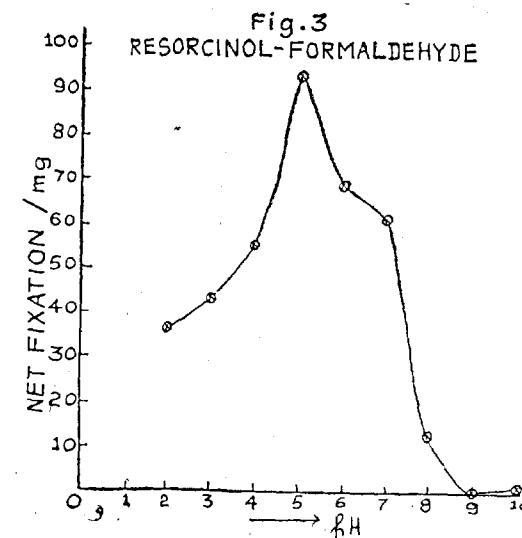


TABLE VI
160 mg. resorcinol / 20 hours / 2 g. collagen.

pH	Amount of resorcinol used up / mg.	
	Blank	Collagen
2	67.6	103.60
3	10.3	65.92
4	10.3	65.92
5	6.7	100.30
6	44.1	112.45
7	93.56	154.57
8	144.21	156.38
9	156.88	156.38
10	155.07	156.38

The reaction of resorcinol and bis hydroxymethylurea with collagen: Stability of resorcinol-bis hydroxymethylurea mixture in aqueous medium :

Bis hydroxymethylurea was prepared as recommended by Walker.¹¹ The crude product was crystallised from 80% ethanol; m.p. 127°C (lit.¹⁴ 126°C.).

A solution prepared from 110 mg. of resorcinol, 120 mg. of bis hydroxymethylurea and 120 mg. of sodium chloride was adjusted to pH and allowed to stand at room temperature for 20 hours. The above experiment was repeated using same amounts of reactants but from pH 4 — 10. The residual resorcinol was estimated in the usual fashion.

The blank experiment was repeated using 2 g. lots of collagen. After allowing to stand for 20 hours the mixture was filtered, the collagen washed repeatedly with water, the total volumes made up and resorcinol estimated.

TABLE VII

110 mg. resorcinol; 20 hours; 2 g. collagen

pH	resorcinol used up/mg.	
	Blank	Collagen
3	.. 101.0	—
4	.. 84.87	20.26
5	.. 77.70	16.67
6	.. 9.25	20.26
7	.. 7.7	25.65
8	.. 13.08	31.02
9	.. 20.26	32.87
10	.. 32.82	57.95

Reaction of phenol-hexamethylene tetramine complex with collagen: The stability of phenol-hexamethylene tetramine complex in aqueous medium :

The reaction of resorcinol-hexamethylene tetramine complex was repeated using 200 mg. of phenol-hexamethylene tetramine complex $[3 \cdot C_6H_5O(CH_2)_6N_4]$. After allowing to stand for 40 hours the mixtures were filtered, the collagen washed repeatedly with water, the total volumes made up and phenol estimated by the bromide-bromate method. A blank experiment was also made using 200 mg. of the complex. The net fixation was plotted against pH; results are presented in Figure I.

TABLE VIII

200 mg. complex/40 hours/2 g. collagen

pH	Amount of complex used up/mg.	
	Blank	Collagen
3	.. 11.6	15.7
4	.. 9.4	18.0
5	.. 13.9	22.6
6	.. 11.6	18.0
7	.. 9.4	18.0
8	.. 13.9	15.7
9	.. 11.6	15.7
10	.. 11.6	20.3

Reaction of phenol-hexamethylene tetramine complex and bis hydroxymethylurea with collagen: Stability of phenol-hexamethylene tetramine, bis hydroxymethylurea mixture in aqueous medium :

The reaction of resorcinol and bis hydroxymethylurea with collagen was repeated using 140 mg. of complex and

120 mg. of bis hydroxymethylurea. A blank experiment was also carried out in the usual manner.

TABLE IX
140 mg. complex/40 hours/2 g. collagen

pH	Complex used up/mg.	
	Blank	Collagen
3	.. 9.1	12.2
4	.. 11.4	9.7
5	.. 11.4	14.9
6	.. 9.1	17.2
7	.. 13.6	12.2
8	.. 11.4	12.2
9	.. 9.1	14.9
10	.. 9.1	12.2

Reaction of catechol-hexamethylene tetramine complex with collagen: Stability of catechol-hexamethylene tetramine complex in aqueous medium:

The above experiments were carried out as described in the case of resorcinol-hexamethylene tetramine complex.

TABLE X
200 mg. complex/40 hours/2 g. collagen

pH	Complex used up/mg.	
	Blank	Collagen
3	.. 17.5	17.5
4	.. 15.0	40.0
5	.. 15.0	15.0
6	.. 15.0	40.0
7	.. 40.0	40.0
8	.. 15.0	40.0
9	} Reaction mixture darkened and estimation proved impossible	
10		

Discussion

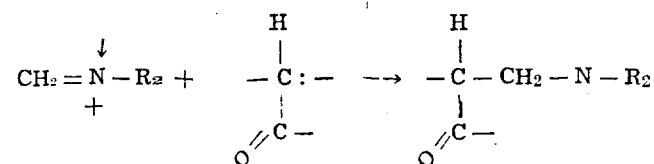
The resorcinol-hexamethylene tetramine complex was found to be quite stable from pH 3 — 8 and shows tendency for decomposition above pH 8. This tendency is exhibited in a more pronounced manner in the case of resorcinol formaldehyde mixture. An equimolecular mixture of bis hydroxymethylurea and resorcinol followed the pattern of resorcinol-hexamethylene tetramine complex but the decomposition seemed to be very great at pH levels from 3 — 6. Catechol-hexamethylene tetramine complex exhibited an almost uniform tendency for decomposition throughout the pH range. The phenol-hexamethylene tetramine complex was found to be quite stable throughout the entire pH range, even when admixed with bis hydroxymethylurea. It seems clear that the stability of the complexes or mixtures depended to a large extent on the nature of the formaldehyde donor and on the reactivity of the phenol.

In the case of the reaction of resorcinol-hexamethylene tetramine complex with collagen, it is quite evident from Fig. I that the fixation occurs best from pH 3 — 6. This is in accordance with Fraenkel-Conrat's observation⁹ that the Mannich base formation takes place predominantly at neutral or slightly acidic pH conditions in the case of the reaction of gramicidin with formaldehyde and *p*-cresol. From pH 6, the fixation decreases and after a dullness around 9 starts to rise again. This behaviour is also observed in the case of bis hydroxymethylurea and resorcinol and in the reaction of resorcinol complex with deaminised collagen (fig. II). The marked dullness in fixation up to pH 7 in the case of the reaction of deaminised collagen with resorcinol complex shows that amino groups are involved in the reaction.

The fixation of the complex in deaminised collagen bears a close resemblance to the fixation of formaldehyde in deaminised collagen. The fact that there is irreversible

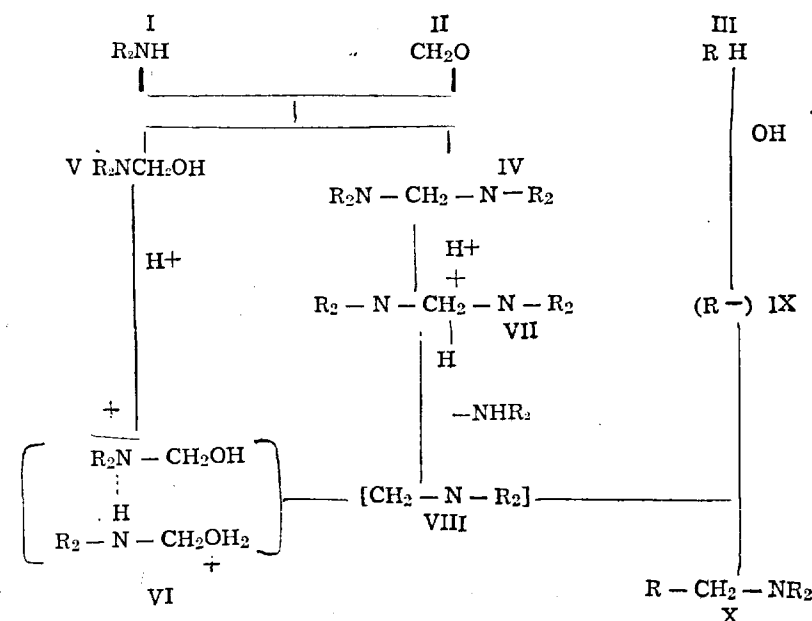
fixation in the alkaline side also indicates probably the formation of chemical bondings. Indeed treatment of the pelt with resorcinol complex at pH 10 produced a leather having a shrinkage temperature of 95°C (Shrinkage temperature of leather produced below pH 7: 98°C.). Of course the tanning was extremely slow and took 15—17 days for completion. Even though no definite conclusions could be drawn regarding the nature of the reaction taking place above pH 7, from the data obtained, it looks as though the guanidino groups are involved. This is not a far fetched possibility since Fraenkel-Conrat and co-workers⁶ have shown that even the guanidino groups are capable of forming cross links with formaldehyde. There is one drawback for such a hypothesis since if it were so the fixation of complex on collagen should have shown a steady upward trend. But this may be due to the variant difficulties that are faced when the pH rises above 7. Even though the amino as well as the guanidino groups are available for reaction above pH 7, the nature of the complex and conditions under those environments are rather inhibitive for formation of Mannich linkages.

According to Lieberman and Wagner¹⁵ the formation of a C-Mannich base involves addition of a methylene ammonium cation or a protonated dialkylaminomethanol, to a carbanion:



As the reaction takes place only in one medium it is quite evident that the reaction involves dual catalysis in an

amphoteric medium. The course of a Mannich reaction could be simply represented as follows:



Excess of acid in the system though favouring the formation of ion VIII checks the formation of IV and V and the carbanion IX while excess of alkali though favouring the formation of IV, V and IX checks the formation of VIII. Lieberman and Wagner¹⁵ by their experiments have vouched for the authenticity of these observations.

In collagen-resorcinol-hexamethylene tetramine complex system, excess acid (high H⁺ concentration) whilst favouring the dissociation of the complex and the formation of the ion [=N-CH₂]⁺ from the ε-amino groups of lysine, will check to some extent the formation of [-NH-CH₂OH] and will prevent the increase in the electron density around the active positions of resorcinol. Excess of alkali though favouring to a great extent the formation of [=N-CH₂-OH] and the increase in electron density around the active positions of resorcinol, checks the dissociation of the complex to resorcinol, formaldehyde and ammonia and also

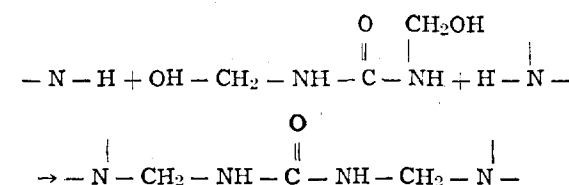
checks the formation of the ion $[=N-CH_2]^+$. As the first step in the formation of Mannich linkages is dissociation of the complex, it is quite conceivable that the reaction can occur only with some difficulty on the alkaline side.

From the reaction of resorcinol with formaldehyde done at various pH levels, carried out according to conditions employed by Windus for tanning it is found that the maximum fixation occurs around pH 5 (Fig. III) where polymerisation is least. This is well in accordance with the principles of the Mannich reaction since at low pH values the reaction should be expected to be hindered by the ready non-availability of free amino groups thus making the formation of amine aldehyde adduct difficult. At higher pH values in addition to the difficulties explained earlier the extreme tendency exhibited for polymerisation of resorcinol and formaldehyde makes the chances of Mannich linkage formation poorer. Windus has done the tanning with the obvious belief that the Mannich reaction takes place only at very low pH value. A study of Fig. II shows clearly that such is not the case. As almost all the resorcinol gets used up in forming polymers at high pH levels, it is difficult to say whether Mannich linkages could be produced above 8. The fact that a leather having a shrinkage temperature of 95°C was produced by carrying out the entire reaction at pH 10 indicates some definite reaction taking place at these pH values. It could also be observed that the polymerisation and fixation reactions are competitive in nature and as the tendency for polymerisation increases, the fixation automatically goes down.

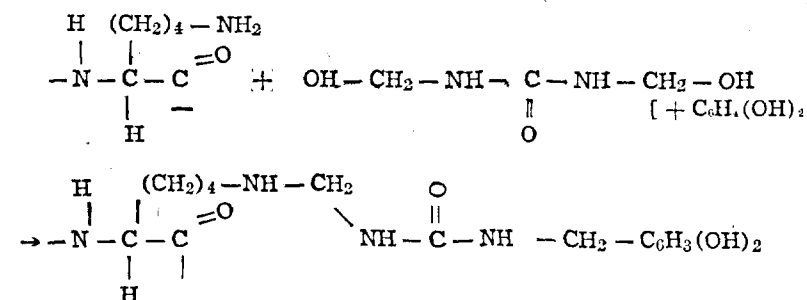
An equimolecular mixture of bis hydroxymethylurea and resorcinol was found to yield a leather having a shrinkage temperature of 87°C , lower than that could be obtained by resorcinol-hexamethylene-tetramine complex but higher than that could be obtained by tanning with bis hydroxymethylurea alone. Bis hydroxymethylurea is known to yield leathers having a shrinkage temperature of 74° and

Gustavson¹⁶ from a study of its reaction with collagen concluded that the compound does not act as a donor of formaldehyde, but fixes as such with collagen forming crosslinks. The fact that on treating the collagen with an equimolecular mixture of resorcinol and bis hydroxymethylurea some resorcinol gets bound irreversibly and the fact that the leathers produced had high shrinkage temperature indicates the formation of C-Mannich linkages.

Bis hydroxymethylurea could be looked upon as an intermediate in the formation of a Mannich base involving urea, formaldehyde and an active hydrogen compound. If tanning with this compound brings about the formation of crosslinks this must take place with the formation of two N-Mannich linkages as follows:



But when resorcinol is present in the medium, bis hydroxymethylurea can condense with resorcinol on one end and with the amino groups of lysine on the other with the simultaneous formation of one N- and one C-Mannich linkages as follows:



This linking could be expected to be stabler than that formed by two N-Mannich base linkages and this also accounts for the higher thermal stability of the leather pro-

duced by employing a mixture of bis hydroxymethylurea and resorcinol.

No conclusive results could be obtained by a study of the reaction of catechol hexamethylene tetramine complex with collagen. The compound did not show any marked fixation but however yielded a leather having a shrinkage temperature of 75°C. The disparity in properties of phenol-hexamethylene tetramine complex and resorcinol-hexamethylene tetramine complex is striking. The phenol complex proved extremely inert.

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15. Lieberman and Wagner, *J. Org. Chem.*, 14, 1001 (1949).
16. Gustavson, *J. A. L. C. A.*, 45, 87 (1950).

RESEARCH BEARS FRUIT

Practical Demonstration No. 7

(Second Series)

Natural and coloured linings from E. I. (vegetable tanned)
hides and skins*

BY

P. S. VENKATACHALAM & T. S. KRISHNAN

Raw materials: Inferior and rejection quality vegetable tanned sheep, goat and calf skins and cow hides that are too defective to be finished into any other type of leather such as shoe upper, suedes, etc.

Lining Kip sides: Vegetable tanned kip sides are taken, weighed, drummed with 800% water on crust weight for 15 minutes, struck out, sammied, split and shaved to 0.9 to 1 mm. substance. The shaved stock is weighed and drummed with 400% water for 20 minutes followed by another wash for 10 minutes. The drum is drained and the sides are ready for bleaching.

Bleaching: A bath is prepared as follows:

Acid oxalic: ½%

Calgon (ICI): ½%

Bleaching syntan

(Basyntan F.C.: 1%

B.A.F.S.)

Water: 200%

The sides are put into the bleaching liquor and drummed for 20-30 minutes, after which a wash with 400% water is

*Process demonstrated during November, 1958.

given for 10 minutes. The goods are taken out, horsed to drain and are ready for retanning.

Retanning: Retanning can be done using either Dhawa (*Anogeissus Latifolia*) leaves or myrobalan nuts.

A porridge is prepared by soaking overnight 30% powdered Dhawa leaves (on crust weight) with water at 50-55°C and the sides are drummed with porridge for 2 hours after which they are rinsed in water, and piled overnight.

In the case of myrobalan retanning, 15% crushed myrobalan nuts on crust weight are soaked overnight with hot water at 50-55°C. More water is added and the nuts are trampled to leach out more tannin and the nuts are removed. The volume of the liquor is made to 100% of the stock. The goods are drummed in this myrob liquor for two hours, rinsed and horsed overnight.

Fatliquoring: The sides are then drummed with 200% hot water at 45°C for 10 minutes after which the water is drained. They are then fatliquored as follows:

Water at 45°C: 150%

T.R.O.: 2¼%

Raw castor oil: ¾%

China Clay: 2%

The hot water is put in the drum and the stock is introduced. The fatliquor emulsion is fed in two instalments at an interval of 10 minutes and the drum is run for 30 minutes after which ¼% formic acid diluted well, is added to the drum in slow feed. The goods are drummed for another 15 minutes after which the leathers are taken out and horsed overnight. In case greater softness is required, the quantity of fatliquor may be increased by 1% keeping the proportion of ingredients the same.

Next day, the sides are struck out, sammed, set well by hand or machine and loosely toggled to dry. The dried

leathers are kept in a pile for 3-4 days after which they are conditioned for moisture, staked, aired off, and trimmed. They are then buffed both on flesh and grain using coarse and fine papers respectively.

Natural Linings: The sides are brushed to remove the buffing dust, measured, cold plated and dusted with French chalk on both sides.

Coloured linings: Usually the linings are finished into pastel shades. The sides are padded on the grain side with a 4% solution of linseed mucilage, dried and cold plated. They are then sprayed with the following pigment coat till they are covered uniformly.

Pigment Coat:

- 1 part of suitable pigment paste
(adjusted to proper shade)
- 1 part of synthetic resin emulsion
- ½ part of 10% casein solution
- ⅛ part of 10% wax emulsion
- 2 parts of water.

Top spraying is done with the following mixture:

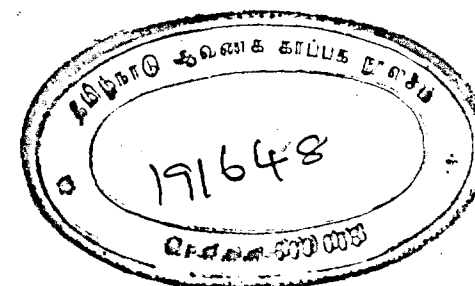
- 5% Casein solution: 1 part.
- 10% formalin: part.

The sides are dried, measured, kissplated and assorted if necessary.

N.B. (a) All formulations are worked out on the shaved weight unless otherwise stated.

(b) Goat and sheep linings are processed similarly.

Handwritten notes in the right margin:
MS 7 on 211, 254
47:5-SSN



TECHNICAL SEMINAR

Liming of Goat Skins for Manufacture of Glazed Kid Leathers*

BY

J. C. DEB

In the manufacture of Glazed kid leather, unhairing is effected by any one of the following methods: (1) Painting with a paste of lime and sodium sulphide and subsequently reliming, (2) using a lime liquor sharpened with sodium sulphide or arsenic sulphide, and (3) enzymatic unhairing. Sodium hydrosulphide and dimethylamine are more in vogue at present, in Western countries. Enzymatic unhairing also is becoming more and more popular. The importance of the unhairing process lies in recovering the valuable by-product, hair, in the least damaged condition.

In the hot climatic conditions prevailing in India, too long a duration of liming would cause loss of hide substance, leading to looseness of grain and poor tensile strength. Hence a series of unhairing experiments have been conducted under various conditions to arrive at a shortened method of liming suitable to the climatic conditions and producing the same unhairing as is obtained by use of sodium hydrosulphide, dimethylamine sulphate, etc. Liming with lime and sodium chloride and sodium sulphide pastes of various compositions have been tried for different durations. Liming in drums also has been studied. The utility of barium sulphide has been investigated.

Steps would have to be taken early for the production of chemicals like dimethylamine sulphate, sodium hydrosulphide in the country, if high quality glazed kid leather is to be produced.

* Summary of the talk given on 10-10-1958.

TECHNICAL SEMINAR

Some points with regard to the Manufacture of Industrial Leathers *

BY

S. N. BOSE

Heavy leather manufacturers are facing difficulties in procuring raw materials needed by them. Correct statistics about the availability of hides for heavy leather manufacture are not available. These should be collected and ways and means should be found to make good the shortage of such hides.

A detailed study of all the factors which control the life of industrial leathers has not yet been made. At present these leathers, for example, the picking bands, are sent to the looms for actual performance. The picking bands prepared at the Institute had a life of 1600-1700 hours. The amount of tannin fixed and its relationship with the life of the picking band leather, duration of sulphur treatment—these are some of the points yet to be studied. Chemical, physical and microscopical studies will have to be undertaken before the effect of these various factors could be assessed.

* Summary of the talk given on 17-10-1958.

CLASSIFIED ABSTRACTS

PROCESS PRIOR TO TANNING

VARIOUS FACTORS RELATING TO QUALITY OF HIDE RAW STOCK FOR TANNERIES. E. J. Strandine and F. G. Connick, *J.A.L.C.A.* 53, 612 (1958). The effects of brining, as compared to ordinary green-salting, have been studied in large-scale curing tests. There is no significant difference in shrink or in leather yield, but brining brings about a more rapid cure, especially if the skins are fleshed first. Brined hides are cleaner than green-salted hides and are almost free from so-called salt stains, salt pits, and raw spots.

THE USE OF SALT TO ASSIST ENZYMATIC UNHAIRING OF FRESH HIDES, WITH HISTOCHEMICAL OBSERVATIONS. Alfred L. Everett and Theone C. Cordon. *J.A.L.C.A.* 53, 548 (1958). Pretreatment with dilute salt solution had been found advantageous in hastening the unhairing of fresh hides with enzyme. A series of controlled tests was performed to elucidate this relationship between salt concentration in the soak solution and the subsequent rate of unhairing, utilizing a numerical scoring system to interpret the data. The effect of frozen storage of fresh hide was also evaluated in the same manner. Results indicated somewhat different concentration optima for salt treatment, depending upon whether or not the hide had been frozen. Among four staining methods applied to thin sections of salt-treated hide the PAS stain successfully demonstrated changes induced in the basement membrane, while a metachromatic stain made other nearby structures visible.

VEGETABLE TANNING

VEGETABLE TANNING WITH ORGANIC SOLVENTS. Yoshiyuki Inoue and Hiroshi Okamura (Kyoto Univ.). *Nippon Hikaku Gijutsu Kyokaishi*, 4, 96-81 (1958); *Chem. Abstr.* 52, 13298 (1958). Fundamental conditions for the tanning by tannic acid, and chestnut, wattle, and SO₂-treated quebracho tannins with the use of MeOH or acetone were examined. In undiluted solvents, it is recommended to treat hide 12 hours in tannin-solvent solution

and to fix 12 hours at pH 4.0—4.6. With higher water dilution rate the tanning rate increased, and the ratios of 3-4 parts solvent and 6-7 parts water were found to be the best. The fixing period could then be 5-6 hours. Examination of the resultant leather indicated that the tanning was homogeneous, but less tannin was fixed in comparison with ordinary tannage. In particular the leather contained less soluble tannins.

TREATMENT OF VEGETABLE-TANNED LEATHER WITH ORGANIC SOLVENTS. Yoshitomi Iizuka and Hiroshi Okamura (Kyoto Univ.). *Nippon Hikaku Gijutsu Kyokaishi* 4, 93-5 (1958); *Chem. Abstr.* 52, 13298 (1958). The amounts of extract from defatted, vegetable-tanned leather decreased in the order of MeOH, acetone, EtOH, dioxane, ethylene glycol, propylene glycol, MeCOEt, and EtOAc. Of the substances extracted by MeOH, tannin was 71.15%; similarly acetone 79.90, dioxane 96.28, and methylketone 94.66. Alcohols generally extracted soluble parts of the tannins in the leather, while dioxane and acetone could extract combined tannins.

AVENS AS TANNIN-BEARING PLANTS. K. F. Blinova. *Sbornik Nauch. Trudov Leningrad. Khim.-Farm. Inst.* 2, 80-90 (1957); *Referat. Zhur. Khim. Biol. Khim.* 1958. *Abstr. No.* 3362; *Chem. Abstr.* 52, 12099 (1958). Tannin content in different organs of *Geum urbanum* and *G. rivale* averaged 12.30% on the basis of dry weight. The tannins of both species belonged to the pyrogallates and contained a small amount of pyrocatechol derivatives. Ellagic acid derivatives were found in the organs of avens of the river variety, and free phloroglucinol was found in all organs except the leaves. Maximum tannin contents were detected in all plants in the spring and in the fall during blooming. The tannin content was lower during the period of fruit formation.

PROCESSES AFTER TANNING

THE CONDITIONS FOR EMBOSSING THE SURFACE OF CHROME LEATHERS. Yu P. Zybin and Z. V. Santalova, *Legkaya Prom.* 18, No. 4, 24-7 (1958); *Chem. Abstr.* 52, 13293 (1958). From studies of the factors affecting the surface finishing of Cr leathers (temperature, moisture content, duration and stress of the pressing, etc.) the following equation is proposed: $H = (1.1T + 87) \cdot (0.0044T + 0.42)$, where H is the impression in μ , T the duration of the pressing in seconds and t the temperature of the press surface. The equation holds between the limits of $t = 60-200^\circ$ and $T = 50-60$ sec. Optimum temperatures lie above 90° ,

pressures should be above 15 kg./sq. cm. and the moisture content not higher than 16%.

TESTING

TANNINS. VII. COLORIMETRIC DETERMINATION OF d-CATECHOL IN CATECHU. Koichi Kimura, Yukishige Satomi, and Shigeaki Kuwano (Univ. Osaka). *Yakugaku Zasshi* **78**, 325-9 (1958); *Chem. Abstr.* **52**, 12439 (1958). d-Catechol (I) was determined colorimetrically with vanillin-HCl reagent. I and gambier tannin were effectively separated by paper chromatography, from which I was eluted out with MeOH and determined. By this means the sum of I and of its optical isomers was obtained and expressed as I value.

COLORIMETRIC DETERMINATION OF THE TANNINS OF HOPS. Jiri Soudek (Ceskoslov. akad. zemedel. ved., Zatec., Czech.). *Sbornik Ceskoslov. Akad. zemedel. ved., Rostlinna vyroba* **3**, 937-43 (1957); *Chem. Abstr.* **52**, 12968 (1958). S. simplified the visual method of de Clerck *et al.* for the determination of the tannins of hops, by use of a photocolorimeter. This eliminates the lengthy visual comparison of the colour extract of hops with a coloured, successively diluted tannin solution. The compensation of the difference in colouration of the primary solutions was modified by calculating the constant K_{tannin} and measuring the extinction E_{tannin} ; for the compensation, an alkalized solution of hop tannins was used. The calculation of the per cent content of hop tannins was simplified. The number of analyses was trebled without loss in precision.

CHANGES IN VISCOSITY OF GELATIN SOLUTIONS WITH TIME. Jaroslav Beneš. *Chem. Průmysl* **7**, 329-30 (1957); *Chem. Abstr.* **52**, 13299 (1958). Measurements conducted at 37° in the Hoeppler viscometer on 56 samples of photographic gelatins over a period of 24 hours showed no correlation between the initial viscosity and its drop with time. The viscosity drop is believed to be the result of several factors, the majority of which are still unknown.

EFFECT OF CONCENTRATION ON VISCOSITY OF GELATIN SOLUTIONS. Jaroslav Beneš. *Chem. Průmysl* **7**, 385-6 (1956); *Chem. Abstr.* **52**, 13299 (1958). The viscosity of gelatin solutions has been studied at concentrations of 2-10% at 35 and 40°, and pressures 10-160 g./sq.cm. Under the given conditions the effect of concentration can be expressed by the following

empirical equation: $\eta = Ae^{kc}$, where η = viscosity c = concentration and A and k are empirical constants.

STRUCTURAL VISCOSITY OF GELATIN SOLUTIONS. Jaroslav Beneš. *Chem. Průmysl*, **7**, 271-3 (1957); *Chem. Abstr.* **52**, 13298 (1958). The influence of pressure on the viscosity of gelatin solutions has been measured in Hoeppler rheoviscometer in the pressure range 10-160 g./sq. cm. at 25, 35 and 45°, and at concentrations of 2, 4, 6, 8, and 10%. The viscosity determination is an important test for the evaluation of photographic gelatins; however, it does not give a complete picture of their actual physical quality in the preparation of a sensitive photographic film. The results are influenced by the test conditions.

DETERMINATION OF AMINO ACIDS BY RADIOISOTOPE DILUTION METHOD COMBINED WITH PAPER CHROMATOGRAPHY. II. AMINO ACID CONTENT OF PROTEINS. Tsuru Wada (Univ. Tokyo). *Nippon Nogei-Kagaku Kaishi* **31**, 743-8 (1957); *Chem. Abstr.* **52**, 12969 (1958). By using the method described in the previous paper, contents of amino acids in several proteins were determined. Taka-amylase, ovalbumin, insulin from whale, and insulin from bonito were hydrolyzed, and the amounts of 8 amino acids were determined. The data were comparable with the data obtained by the other methods. The superiority of this method to the other methods was that it required only 0.5 mg. of protein as the material, and the analytical procedure was relatively simple.

DETERMINATION OF LIGNOSULFONIC ACIDS WITH THE AID OF CHLORINE WATER. Jerzy Gańczarczyk (Inst. Gospodarki Komunalnej, Gliwice, Poland). *Chem. Anal. (Warsaw)* **2**, 441-51 (1957) (English and Russian summaries); *Chem. Abstr.* **52**, 13252 (1958). A new colorimetric method for determining lignosulfonic acids (I) in water is based on the colour reaction of I with Cl water. To 100 ml. of solution, (pH 6.5-7.5) containing I in amounts less than 100 mg./l., add, mix thoroughly, and after of Cl water (containing 5 mg. Cl/ml.), mix thoroughly, and after 15 minutes add 0.2 ml. of NH_4OH . Then determine the colour intensity in mg. of Pt/l. by comparing with standards. This method can be used to determine I in solutions of spent sulfite liquors in concentration from 0.1 to 1.0 ml./l of solution. The factor for recalculating colour intensity (in mg. of Pt/l) into concentration of I in g./l. was $0.213 \pm 5.2\%$ for different spent sulfite liquors. That value was the mean value computed for 10

specimens of various lignosulfonic acid spent liquors. The analogical factors calculated for lignosulfonic acid liquor concentrates and for Howard precipitate were much higher, namely, 0.385 and 0.526 respectively. On this basis it was suggested that I easily underwent transformation during treatment. The studies of α , β_1 , and β_2 fractions by the Cl method showed the same increase of the colour after reaction for all α fractions, a smaller one for β_1 fractions and a decrease of the original colour for β_2 fractions. It seems that the β_2 fraction has no lignin character, whereas the β_1 has it only partially.

CHROMATOGRAPHY OF THE SULFONATED PRODUCTS OF PHENOL-FORMALDEHYDE CONDENSATION. I. PAPER CHROMATOGRAPHY OF PHENOLSULFONIC ACIDS. *Eduard Muck, Drahoslav Kokes, and Ferdinand Langmaier (Vyzkumny ustav Kozedelny, Otrokovice, Czech.). Chem. Listy, 52, 746-8 (1958); Chem. Abstr. 52, 13295 (1958).* Synthetic tannin materials are chromatographed in the systems iso-PrOH-NH₄OH (2:1) and isoamyl alcohol-EtOH-NH₄OH (5:3:2.5) and detected with 1% alc. solution of FeCl₃ or diazonium salts or by heating the spots to 180°. Ultraviolet spectra are charted of the NH₄ salts of 1-hydroxy-2-benzenesulfonic acid (I) and 1-hydroxy-4-benzenesulfonic acid (II), and a chromatogram is shown of a phenol-HCHO condensate constituents. The R_f values on Whatman paper No. 3 in the given systems are resp.: I 0.77 and 0.47 and II 0.51 and 0.19.

THEORIES

HYDROGEN-BOND FORMATION. VII. REACTIONS BETWEEN CARBOHYDRATES AND PROTEINS IN WATER AND THEIR RELATION TO TRANSFER OF CARBOHYDRATES ACROSS RED-CELL MEMBRANES. I. *C. L. Bruce, C. H. Giles, and S. K. Jain (Roy. Coll. Sci. Technol., Glasgow, Scot.). J. Chem. Soc. 1958, 1610-13; Chem. Abstr. 52, 11984 (1958).* By the refractometric method AcH, isobutyraldehyde, terephthalaldehyde, D-glucose, or sucrose fail to show complex formation in H₂O with MeCONHMe, PhOH, *p*-MeC₆H₄OH, casein, edestin, or gelatin; meso-inositol (and simple alcohols) do form H-bond complexes with the protein-type components. It is suggested that normal carbohydrates do not form H-bond complexes with red-cell membranes because of H₂O shielding and, therefore, traverse them more readily than compounds which complex with the membrane proteins.

ELASTIN. II. MUCOPOLYSACCHARIDES IN ELASTIN FIBERS. *Lorenzo Gotte and Vittorio Moret (Univ. Padua, Italy). Atti accad. naz. Lincei Rend. Classe sci. fis. mat. nat. 23, 281-6 (1957); Chem. Abstr. 52, 12940 (1958).* Treatment of lipide-free bovine ligamentum nuchae (yellow elastic connective tissue) with 0.1 N NaOH solubilized most mucopolysaccharide (hexosamine) material (I) along with collagen. The remaining I was firmly bound to the elastin residue (II) since treatment with CaCl₂ at various pH effected no separation. Hydrolysis of II with 5 N HCl for 6 hours gave a positive test for SO₄²⁻ in the product, indicating the presence of chondroitinsulfuric acid in II. Microscopic examination of the periodateleucofuchsin-stained II showed the structural heterogeneity of the sample.

REACTIONS BETWEEN CHONDROMUCOID AND PROCOLLAGEN. *S. M. Bychkov and V. A. Fomina (Lab. Department Health, U.S.S.R., Moscow). Voprosy Med. Khim., 4, 59-64 (1958); Chem. Abstr. 52, 12030 (1958).* Chondromucoid was isolated from the trachea of cattle according to a modification of the method of Shatton and Schubert. Procollagen was prepared from the skin of white rats according to Tustanovskii. After 2 recrystallisations it had 16.1% N, less than 0.1% hexosamine and tyrosine and no SO₄. The method of Alicino was used for the determinations of sulfates and the method of Folin and Marenzi for tyrosine. Procollagen was dissolved in 0.1 N acetate buffer at pH 3.5, chondromucoid in distilled water. On mixing together these solution, the precipitate was formed immediately. Irrespective of the excess of either of the components of this complex, composition did not vary after reaching a certain level. The formed complex of chondromucoid and procollagen is insoluble in water, gradually swells in 0.05-0.5 M NaOH and is soluble in 0.2-0.3 M CaCl₂. It has 12.75% N; 0.60-0.73% tyrosine and 0.93-1.38% SO₄. Its S/tyrosine ratio was 1.50-2.25 and S/N 0.07-0.1. These ratios in chondromucoid were S/tyrosine 3.11 and S/N 0.80.

STUDIES ON THE SPECIFICITY OF COLLAGENASE. *Susan Michaels, Paul M. Gallop, Sam Seifter and Edward Meilman, Biochim. Biophys. Acta. 29, 450 (1958).* When purified collagenase was incubated with highly purified fish collagen (ichthyocol) or calf procollagen or with the gelatins derived from these, the end products were found to have an average molecular weight of about 500. Two dimensional chromatography showed at least 30 peptides to be present at the end of the reaction. The N-terminal amino

acids of these different peptides were determined by the Sangers' technique and only DNP-glycine could be identified in each peptide. The evidence is taken as fairly conclusive to suggest that collagenase acts on collagen to release only N-terminal glycine. About 60% of the bonds involving glycine were ruptured. A reaction mixture of ichthyocol and collagenase was analysed for C-terminal amino acids by the hydrazine method and a number of C-terminal amino acids were identified. Fish gelatin unreacted with collagenase showed only the presence of C-terminal glycine and alanine of which glycine was predominant. Quantitative determination of C-terminal glycine in both cases showed that the amount did not increase after enzymic action which indicate that collagenase does not cause the release of C-terminal glycine. It is therefore suggested that either the sequence of glycyl-glycine cannot be split by the enzyme or it does not occur in sufficient frequency in the protein to be detected. Since approximately 60% of the peptide bonds involving glycine are ruptured with little regard to the nature of the preceding amino acid residue it is suggested that a larger sequence of amino acid residues may contribute to the specificity of the enzyme course of hydroxylation of lysine to form hydroxy lysine in collagen.

K. T. J.

DEGRADATION OF COLLAGEN BY HEAT: James M. Cassel, *J.A.L.C.A.* 53, 507 (1958). Purified steerhide collagen heated at temperatures ranging from 125° to 170°C in vacuum, air, and oxygen for periods of 3 days shows reduced capacity to absorb water vapor and increased solubility in water. Small changes are apparent in the infrared spectra of the most severely degraded materials. Losses in the amounts of a number of amino acids as the temperature of heating is increased are shown by quantitative paper-chromatographic analysis. In particular, methionine, serine, threonine and tyrosine are affected. Collagen heated in air at 100°C for 3 days at relative humidities ranging from 20% to 90% shows increased solubility in water but relatively little change in amino acid composition. The water-soluble fractions obtained by heating at high relative humidities are of somewhat higher molecular weight than those obtained by heating in the absence of moisture.

STRUCTURE OF PROTEINS. V. PROGRESSIVE DEGRADATION OF THE POLYPEPTIDE CHAIN FROM THE N-TERMINAL AMINO ACID. Luciano Scarso, Ernesto Scoffone,

and Doroteo Chellemi Univ. Padua, Italy). *Gazz. Chim. ital.* 87, 1348-55 (1957); *Chem. Abstr.* 52, 13813 (1958). Selective reduction of N-(2, 4-dinitrophenyl) peptides (I), and lactamization of the resulting N-(2-amino-4-nitrophenyl) peptides (II) gave 3-substituted 3, 4-dihydro-2-hydroxy-7-nitroquinoxalines (III) with simultaneous liberation of a degraded peptide suitable for coupling with 2, 4-(O₂N)₂C₆H₃F and repetition of the degradation cycle. I (5.4×10^{-5} mole) prepared according to Sanger, in 2 ml. 95% alcohol was treated with 2 ml. (NH₄)₂S, prepared by saturating 8N NH₄OH with H₂S; the mixture was heated 6 hours at 37° in a sealed ampul, diluted with 95% alcohol to about 45 ml. and acidified with 6 N HCl to pH 2.2 (glass electrode), the volume adjusted to 50 ml. with alcohol and the product lactamized 6 hours at 37° in a sealed ampul. Samples of the solution (0.1 ml.) spotted on Whatman No. 1 paper were chromatographed in 0.1 M acetate buffer (pH 5.5) and the spots of III eluted with 95% EtOH and evaluated spectrophotometrically. The solution was concentrated *in vacuo* and extracted repeatedly with EtOAc saturated with H₂O, the aqueous phase neutralised with NaOH; to this solution NaHCO₃ was added to a concentration of 7%, the mixture treated 3 hours at room temperature with 0.2 g. 2, 4-(O₂N)₂C₆H₃F in 10 ml. Me₂CO and evaporated *in vacuo*, the aqueous solution extracted 5 times with 5 ml. Et₂O and acidified with N HCl to pH 1, extracted 3 times with 10 ml. EtOAc, the extract evaporated *in vacuo*, the residue chromatographed on Celite 545 buffered at pH 7.25 with phosphate buffer and the column eluted with CHCl₃ saturated with buffer and CHCl₃ saturated with 1:3 buffer-AcEt to give the secondary I. The eluate was evaporated *in vacuo* and the residue reduced, lactamized as above, and the 2nd amino acid determined as III. Tabulation showed a notable difference in the formation of III from glycine and leucine peptides though the adjacent amino acid exercised no particular influence. No destruction of III takes place at pH 2.2 at 37° in 6 hours but the rate of cleavage is dependent in the terminal group. Lactamization at 55° in 6 and 24 hours raised the yields of III from glycyltryptophan to 69.5 and 76.5% respectively. Optimum reductive cleavage and lactamization at pH 2.2 were obtained in 6 hours at 37°. The degradation procedure applied to lysozyme, trypsin, and cattle serum albumin recovered —, 0.71, and 0.75 mole, respectively of the N-terminal lysine, isoleucine, and aspartic acid groups, respectively. The estimation of the yield of lysine from lysozyme was hindered by ignorance of the molar extinction coefficient of III for lysine.

COLLAGEN. XII. ACTION OF DIFFERENT POLYOSIDES (INCLUDING BACTERIAL POLYOSIDES) ON NATIVE COLLAGEN (TENDON COLLAGEN). Suzanne Bazin, Albert Delaunay, and Nicole Briquet (Inst. Pasteur, Garches, France). *Ann. Inst. Pasteur*, 94, 7-14 (1958) (English Summary); *Chem. Abstr.* 52, 13816 (1958)—Shredded rat-tail tendons were macerated for 48 hours at 4° in a 3.3M KCl solution at pH 7, washed successively with H₂O and 0.1M NaCl at pH 4.3, left 48 hours in the latter, and finally washed with H₂O. Tendons so treated, although macroscopically normal, swelled in 48 hours in 0.15 or 0.427M NaCl at pH 7 and their collagen was dissolved. But in the presence of the mucopolysaccharides (heparin, chondroitinsulfuric acid) or bacterial polysaccharides (typhoid lipopoly saccharide C_x, pneumococcus polysaccharide C, streptococcus polysaccharide, staphylococcus polysaccharide) similarly prepared tendons swelled slightly or not at all. The effect of mixtures of these substances was cumulative. The tendons swelled in the presence of S II pneumococcus polysaccharide, inulin, glycogen, and soluble starch.

CHEMICAL MODIFICATION OF THE SIDE CHAINS OF GELATIN. A. W. Kenchington (Brit. Gelatine & Glue Research Association, London), *Biochem. J.* 68, 458-68 (1958); *Chem. Abstr.* 52, 11984 (1958). The ionizable groups in gelatin have been modified chemically in order to observe any resultant effects on the solution viscosity and gel rigidity. No effect not attributable to degradation was detected. Complete esterification of COOH groups was achieved, since the analytical figures for methyl content agreed with the total free COOH content before treatment. Highly esterified material was unstable in aqueous solution at all pH values. Complete-N-acetylation was achieved by the action of Ac₂O on gelatin dissolved aqueous AcONa. Little O-acetylation occurred until the majority of the amino-groups were acetylated. The O-acetyl groups were easily removed by mild alkaline treatment. Guanidation of amino groups was shown to proceed virtually to completion. Under all conditions of reaction studied, hypobromite was found to produce extensive degradation and other changes before its reaction with guanidino groups was complete.

ACTION OF PERIODATE ON COLLAGEN AND KERATIN. Helmut Zahn and Ludwig Zurn (Univ. Heidelberg. Ger.). *Z. Naturforsch.* 12b, 788-91 (1957); *Chem. Abstr.* 52, 13813 (1958). Hydroxylysine incorporated in tendon collagen was oxidized with 0.24 M NaIO₄ solution at 40° for 72 hours. Tyrosine, cystine, and methionine as well as wool were also attacked. The periodate

reaction can thus not be regarded a specific reaction for carbohydrate.

CHEMICAL STUDIES ON THE HAIR. I. SULFUR-CONTAINING AMINO ACIDS ESPECIALLY ON LANTHIONINE. Masaru Funatsu and Motosuke Kikutani (Kyushu Univ. Fukuoka). *Nippon Nogei-Kagaku Kaishi* 30, 572-6 (1956); *Chem. Amstr.* 52, 12000 (1958). Changes in the total S, alkali-labile S, and cystine S were established after boiling the hair with 0.2% Na₂CO₃ for 30-60 minutes. Untreated hair contained total S 5.24, alkali-labile S 5.06, and cystine S 3.58%. Thus the majority of S was alkali-labile, and a part of which was stable in Na₂CO₃ treatment. Cystine was almost destroyed by Na₂CO₃, and a part of cystine was converted into lanthionine (I). The alkali-labile S which was resistant to Na₂CO₃ treatment was not cystine S, and the amount of this fraction coincided with S fraction which formed SO₄= by Br oxidation. This fraction was considered to be I, and it was suggested that I was present in hair as a normal amino acid. **II. MELANIN OF DARK HUMAN HAIR. 1. PREPARATION OF MELANIN FROM HAIR.** Masaru Funatsu and Gunki Funatsu. *Ibid.* 31, 221-5 (1957) Melanin (II) was prepared from black hair by 2 methods: (1) hair was treated with concentrated H₂SO₄ at 33° for 12 days, and the extract diluted to precipitate II (named S-II); (2) hair was soaked in N NaOH at 30° for 48 hours, and black residue was washed with water to give B-II. The II was composed of a protein (III) and pigment; a large amount of Fe was present in II together with small amounts of Mg, Al, Sn, Zn, Cu, Ni, and Ti. The III portion consisted of 14 amino acids; the ratio of lysine and arginine in it was 1:2. It was stated that the hair was a compound III composed of leucokeratin and melano-protein. **III. OXIDATION OF HAIR BY HYDROGEN PEROXIDE.** *Ibid.* 874-8. Dark hair was treated with 30% H₂O₂ to decomposition II; the residue of decolorized hair III consisted of 4 components when examined with ultracentrifuge and electrophoresis; the major component was a fibrous III, soluble in 0.1 M Na₂CO₃-NaHCO₃ buffer pH 9.8, molecular weight 30,000, and axial ratio about 20. The III portion of II was different from fission products of partially oxidized keratin.

PATENTS

GLYCEROL POLYGLYCIDYL ETHERS AS TANNING AGENTS. Edward M. Filachione and Edward H. Harris, Jr. (to the United States of America, as represented by the Secy. of Agr.) U.S. 2,829,943, Apr. 8, 1958; *Chem. Abstr.* 52, 12439 (1958). Glycerol polyglycidyl ethers (I), of the general formula,

$(\text{OCH}_2\text{CHCH}_2\text{O})_x \text{R} [\text{OCH}_2\text{CH}(\text{CH}_2\text{Cl})_y \text{OCH}_2\text{CHCH}_2\text{O}]_z$ in which R is a glyceryl residue, x, y and z are 1-2, and $x+z$ is 2-3, are used preferably in aqueous alkaline suspension for tannage of skins and hides. Thus, pickled cowhides were depickled in 5% aqueous NaHCO_3 and a portion equivalent to 15 g. (dry wt.) of the wet hide was milled with a suspension of 10 g. of a I ($x=y=z=1$) in 200 ml. of water. During 2-day tannage, the pH fell from 8.1 to 6.9 and the shrinkage temperature increased to 80°. The leather was firm, white, flexible and of high quality, with special ability to retain its leathery character even when heated above its shrinkage temperature. Examples describe the application of the process to both cowhide and calfskin with such agents as Na lactate, NaHCO_3 , Na_2CO_3 , MgO, and $\text{Ca}(\text{OH})_2$ to control the pH. Cowhide requires one or more days for thorough tannage, while calfskin tannages are complete in 8 hours. Retannage with vegetable extract is possible in the conventional manner, bisulfited quebracho and chestnut extracts being used during a 4-day retannage. The shrinkage temperature increased during retannage from 85 to 91° and the weight gain was 50%.

TANNING WITH ACID SOLUTIONS OF ALKALI METAL ZIRCONIUM SILICATES. Wm. J. Rau and Ian C. Somerville (to Rohm & Haas Co.). U.S. 2826,477, Mar. 11, 1958; *Chem. Abstr.* 52, 12439 (1958). Water-insoluble double silicates of Zr and alkali metals are used to tan pickled skins, producing leathers with equal or superior plumpness, mellowness, and fullness to those produced from water-soluble Zr salts in the absence of silicic acid. The double silicates include those in which the molar ratio of $\text{SiO}_2:\text{ZrO}_2$ is $\leq 1.5:1$, preferably about 1:1 and in which the ratio of SiO_2 to the oxide of the metal is $\leq 3:1$ preferably about 1:1 compounds having the formulas $\text{Na}_2\text{ZrSiO}_5$ and $\text{Na}_4\text{Zr}_2\text{Si}_3\text{O}_{12}$ fall within the range of usable compounds, the former being preferred.

TANNING MATERIALS GIVING WHITE LEATHER. Cassella Farbwerke Mainkur Akt.-Ges. (Karl Keller, inventor). Ger. 892,817, Oct. 12, 1953; *Chem. Abstr.* 52, 12440 (1958). Skins treated with a water-soluble condensation product of an amino compound of a diazine, triazine, or triazole (etherified with a lower alcohol if required) with a methoxyaldehyde give white, soft, but abrasion-resistant leather. These tannins do not possess the un-

desirable characteristics (poor water solubility and decomposition with evolution of HCHO in hot water) of melamine HCHO condensation products used hitherto. As a 1st component, melamine (I), its phenyl, alkyl or acyl derivatives, trihydrazinotriazines, alkyl ethers of ammeline, or thioammeline, guanamines, aminodiazines, hydroxyaminodiazines, aminoazoles, or guanazoles are used. These are condensed with alkoxyaldehydes, such as trimethyl (hydroxy) acetaldehyde (II), bis(hydroxymethyl) propyl aldehyde, or hydroxymethylisobutyl aldehyde. Other aldehydes, e.g., HCHO, or substances condensing with aldehydes, e.g. urea, can be added. The crystalline or sirupy products are very soluble in water at normal temperatures, giving almost odorless, clear stable solutions. If the tanned leather has a yellow tinge, a blue optical bleach can be added. Thus, a mixture of 31.5 parts I with 124 parts 81% aqueous II was made alkaline with approx. 0.2 part 10N NaOH, stirred, and warmed gradually to 75°, a further 0.4 part NaOH solution being added meanwhile. The clear hot solution of the condensation product was then stirred with 315 parts MeOH, cooled, and acidified with 20 parts 10N HCl at 18-20°, stirred a further 30 minutes, and neutralised with soda. After filtering, the MeOH was evaporated at 45° *in vacuo*. The sirupy residue was filtered free of inorganic salts or used directly. In another example, acetoguanamine 62.5 was dissolved in aqueous II 233 and water 75 at 75-80°, and MeOH 600 and oxalic acid 0.8 part added. The mixture was stirred and refluxed gently for 30 minutes, cooled, neutralised with soda and filtered. The MeOH was evaporated and the residue concentrated to constant weight in an air stream at 45-50°. The clear sirup remaining was highly miscible with water. A tanning procedure using these reagents is also described.

BYE PRODUCTS

THE ADDITION OF HYDROTROPIC-ACTIVE AGENT IN THE PRODUCTION OF ANIMAL GLUE AND GELATIN. G. Alexandrescu and A. Süsser. *Rev. Chim. (Bucharest)* 7 360-7 (1956); *Chem. Abstr.* 52, 12439 (1958). In the production of animal glue, the pretreatment of the washed hide substance with lime was changed to a short treatment with hydrotropic active substances, thus shortening the production process by approximately 60 days. Active hydrotropic materials tested were: HCl (at equil. pH 2); 1% and 0.1% CaCl_2 solutions; 1% and 0.3% urea solutions; 1.0, 0.3 and 0.1% solutions of acetamide; ZnCl_2 ,

0.1% solution (ineffective); and NH_4CNS 0.1% (red colour with Fe). The hide material is washed 24 hours, delimed with a 2% solution of NH_4Cl , washed thoroughly, and brought in contact with the aqueous solutions of the hydrotropic agent for 24 hours. The hides were taken to the cooking process without washing, except when CaCl_2 was used. Best results were obtained with 1% CaCl_2 or 1% acetamide solution at pH 6, while the optimal temperature range was 50-9°. A production test on industrial scale showed a 70% gain in yield by this method vs. the classical lime process.

GENERAL

CHALLENGE OF THE SPACE AGE. *Anon. Leather & Shoes*, 135, No. 25, 8 (1958). The U.S. Leather and Leather Products Industry, if alert to the possibilities and probabilities of the space age can play an important role in it. Because leather itself contains so many desirable properties, the industry has a chance to supply various materials. Further the industries arising with the space age will find leather adaptable and even necessary. As a matter of fact, leather has already found a place in space travel. The space suit recently developed uses leather in the special boots, gloves and at the elbow where extra wear and flexibility of movement is required. Scientists believe that leather, because of its skin like properties, its ability to withstand extremely low temperature without becoming rigid, its qualities of comfort, durability and flexibility, can be an important part of space suiting.

The Navy also has used leather throughout the interiors of its atomic powered submarines. Upholstery leather has been found to have a high degree of resistance to radiation contamination. Once exposed, it can be easily decontaminated by washing. It has been found most suitable due to its resistance to wear and abrasion. Special clothing developments, in connection with launching space vehicles include moulded sole leather boot, a new type leather glove which may be dipcoated to protect the wearer against extreme temperatures and radiation and a leather soling, tanned in a fraction of the time conventionally required.

Chrome tanned cowhide type leather, with most of the excess oils and greases removed, would be about 25% less dense than conventional leather and would be valuable for space apparel like boots, gloves, jackets, linings, etc., and also for upholstery, wall

coverings in space ship and space station interiors, control coverings, bailout equipment parts, food containers, etc.

A light weight sheep skin leather, which can be dipcoated with suitable compositions, and which retains flexibility and provides protection against liquid fuels, acids, radiation, cold and heat, has been developed; it is comparatively simple to produce, inexpensive and durable.

The Army's new tropical boot, with a vulcanised bottom and treated leather upper, is resistant to radiation, does not shrink under dry heat and resists wet heat through water repellancy.

Technologists believe that leather will be one of the materials that will enable man to reach the moon, explore it and return to earth.

V. S. P.

LEATHER IN THE SPACE AGE. C. W. Mann. *Leather & Shoes*, 135, No. 25, 21 (1958). In spite of its being well-known, leather may not be considered in the search for materials for use under extreme cold conditions, for the reason that its low temperature characteristics have not been explored. Leather retains at -300°F most of the physical properties, that it has at ordinary temperature. The softness and flexibility of leather stem from its fibrous structure. Any possible stiffness of leather at low temperatures would be due to fillers, salt deposits, fats and oils. It has been shown that fatliquored chrome leather, after degreasing, remains pliable and is ideal for arctic ski bindings and rucksack straps, the removal of oil, improving serviceability over a much wider temperature range and not lowering the flexibility. Leather could possibly be made with about 1/10 the oil used at present.

Toughness, thermal expansion and contraction, and stretch remain practically the same between -94°F and 158°F and probably will remain so at still lower temperatures. It is difficult to impart these characteristics to synthetics and hence leather should become even more important with rapid expansion of our knowledge about it.

V. S. P.

NEW VISTAS FOR LEATHER AHEAD. *Fred O'Flaherty, Leather & Shoes*. 135, No. 25, 28 (1958). The challenge of producing a space age leather could be met, if we consider some of the recent researches. The polar groups have been considered as active spots for tannin combination. With better knowledge of the terminal groups of collagen, and its partial hydrolysis products, i.e., of the chemical architecture of collagen, it would be possible to replace one or more amino acids with some metallic body and thus make a leather absorbing unwanted radiations. The study of reactivity of collagen with metals could give such information. Newer lubricants and fungicides will be of great value in rendering the new leather suitable for its new purpose. Leather resistant to X-rays penetration has been provided by the Industry.

V. S. P.

NOTES & NEWS

PROF. HUMAYUN KABIR DECLARES OPEN THE NEW C.L.R.I. WORKSHOP BUILDINGS

The new Workshop buildings of the Central Leather Research Institute were declared open by Prof. Humayun Kabir, Vice-President, Council of Scientific & Industrial Research, and Minister for Scientific Research and Cultural Affairs, Union Government, on Friday, the 24th October, 1958, at 10-15 a.m.

Welcoming the Minister and the gathering, Dr. Y. Nayudamma, Director of the Institute said, that the construction was taken up in March, 1958 and had been finished fairly ahead of schedule. The American Technical Co-operation Mission had generously given several of the machines in the Workshop and more were expected later. The Workshop was intended for the repair, upkeep and maintenance of the various machines and tools used in the Institute and also for fabrication of machinery needed by the small-scale producer.

Prof. Humayun Kabir said that it was always a pleasure for him to initiate new activities in the National Laboratories. He expressed his gratification at the fact that this Workshop was intended not only for maintenance of equipment but also for designing and fabricating new machines. In view of the difficult foreign exchange situation and in view of the vast national reconstruction, machinery needed would have to be designed and fabricated, when-ever new projects or new lines of activity were taken up. In a planned industry, like Leather and Leather Goods Industries with small-scale units, devising of such equipments as would suit the small-scale units would increase the scope for employment and he would be very happy to find how this Workshop helped in that direction. In this connection, he emphasized that everyone working in the Institute was a scientific worker because he was contributing to the total scientific progress.

Prof. Kabir next referred to the special significance of the Leather Institute, for in this country not only the trade of tanning, but also the people engaged in it, had been looked down upon.

So this Institute should help in making the people realise the importance of this trade, which was as honourable and as dignified as any other craft.

Science was a continual revision of what had happened and there was always decentralisation of knowledge in science. Any process developed should not be kept secret but be openly proclaimed so that it could be tested. He would at the same time point out that the Institute had to deal not only with practical application but also the theoretical or fundamental part. Fundamental research advanced the frontier of knowledge that might be useful some time hence. He hoped that this would be the spirit in the Institute.

ASSOCIATESHIP DIPLOMA OF THE INSTITUTION OF CHEMISTS (INDIA)

In a recent Office Memorandum of the Ministry of Scientific Research and Cultural Affairs, the Government of India have decided that the Associateship Diploma of the Institution of Chemists (India), by examination be recognised for all chemical appointments for which M.Sc., degree in Chemistry is prescribed as a qualification.

GOVERNMENT LEATHER INSTITUTE, KANPUR

The Government of Uttar Pradesh have renamed 'The Government Leather Working School', Kanpur as 'Government Leather Institute', Kanpur in view of the increased facilities for higher technical training offered by it, including tanning, after its re-organisation in accordance with the recommendations of the Visiting Committee of the All India Council of Technical Education, Union Ministry of Education.

TREATMENT OF FULLY TANNED LEATHER WITH EPSOM SALT

The treatment of heavy vegetable tanned leather prior to drying with strong solutions of Epsom salt to which additions of sugars are usually made, is frequently practised to improve colour and suppleness. The process is more commonly carried out by drumming than in pits and assists penetration in the absence of sugars.

The presence of Epsom salt causes a salting out or precipitating action on the excess tanning matter of the interstices of the

hide which otherwise might come to the surface during drying and varies with different materials. The use of sugar prevents the formation of larger aggregates which could clog the pores.

This treatment tends to reduce the moisture content in the more open parts of loose hide structure whilst the presence of sugars also aids salt penetration by restraining precipitation and may also lessen any tendency to spue formation. Epsom salt treatment has the further advantage of improving the final colour and reducing any tendency to crackiness should the leather become very dry.

Leather Trades Review, 129, 182 (1958).

CEYLON LEATHER FACTORY TO TREBLE OUTPUT

The Leather Corporation of Ceylon, a Government-sponsored body has plans for a new factory to increase its annual production from 30,000 pairs of shoes to 100,000. A Danish Expert who has arrived in Ceylon and a Czechoslovakian Expert, who is due to arrive next month, will be consulted before the Corporation formulates its plans.

Leather Trades Review, 13-8-1958.

RESEARCH BEARS FRUIT

Practical Demonstration No. 8

(Second Series)

Process: Manufacture of Natural and Waxed Chrome Sole Leather.

Period: 15th December, 1958 to 12th January, 1959.

Place: Central Leather Research Institute, Madras-20.

A process for the manufacture of Natural and Waxed Chrome Sole Leather has been worked out at this Institute. It is proposed to demonstrate this process as the next in the Second Series of Practical Demonstrations from 15th December, 1958 to 12th January, 1959.

If you are interested in this line of work, you are most welcome to send your representative/s to get trained in the manufacture of this type of leather. You may kindly intimate the name/s of the representative/s who is/are likely to attend this demonstration. After following the process, we would request you to try this process in your tannery and give us your valuable comments.

C. L. R. I. NEWS

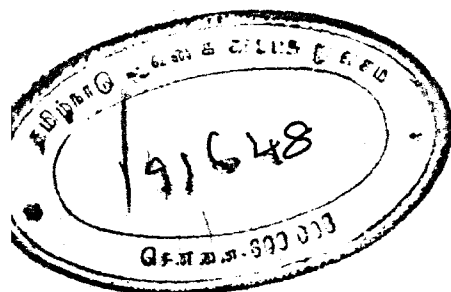
Dr. S. K. Barat, Senior Scientific Officer, Grade I, has been appointed on promotion as Assistant Director, with effect from 16th October, 1958.

Shri S. C. Nandy has been awarded the degree of Master of Science of the Madras University for his thesis "Studies on the Bacteriological and Chemical aspects of the curing of Indian hides and skins."

Shri W. Madhavakrishna has been awarded the degree of Doctor of Philosophy of the Madras University for his thesis "Studies in Enzymic Unhairing with reference to Leather Manufacture".

The first of the Practical Demonstrations by the Central Leather Research Institute at outside centres as part of Extension Service Programme has been started at Jallundur on the 6th October, 1958. The processes to be demonstrated are Vegetable Tannage by the E.I. technique and Rapid Sole Leather Tannage. The demonstrations are being conducted by the Central Leather Research Institute Team headed by Shri A. Ganesan, Senior Scientific Officer.

The Central Leather Research Institute is participating in the 'India-1958' Exhibition, which was inaugurated on the 8th October, 1958, by Shri Jawaharlal Nehru, Prime Minister of India. The C.L.R.I. stall is in the Science and Technology Pavilion.



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For further information please contact

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