

Everything You Wanted to Know About Collagen Models - But Were Too Afraid to Ask! (Part II)

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Abstract

Colloidal Chemistry has developed tremendously in the last century, yet its principles are just not agreeable to the leather industry and are regarded with great mistrust. Some of these colloidal concepts can be very useful to produce better leather, in better ways. Such then should be the ultimate goal of any working chemical model. A series of chemical equilibriae between the tautomers and related conformers of beam-house treated collagen being processed into leather, are proposed. This model includes a micelle analogy of the triple-helix as a thermodynamically akin aggregate to the colloidal micelles formed by surfactants with water. They can help illustrate some of the properties of the collagen conformers, such as the Zwitterion tautomer, in which its charges are stabilized by water, and thus interact with many tannery-used chemicals by means of ionic saline non-covalent bonds at colloidal dimensions. By careful controlled structural destabilizing of collagen, more carboxyl side-groups are effectively more exposed to help exhaust chrome more efficiently. Significant savings of chemicals and intelligent modification of the tannery unit operations have evolved from the application of colloidal concepts; demonstrated in trials by the firm ABC Leder-Grupo *Andino* S.A. Much work is yet to be done at participating tanneries according to what is practical and commercially required. These studies are presently continuing at industrial level, based on the early presentation at the 107th ALCA Congress of 2011 at Red-Wing, MN. USA.

Keywords: Canonical/Zwitterion tautomers, Dehydration/hydration of Peptides, Triple Helix as a Unit for collagen, Proton Exchange processes by means of a water conductor “wire”, non-covalent long-ranged saline bonds.

Introduction

In providing a working chemical model one should specify briefly what the model is not intended to be: This is not a “*unified-field*” model that will succinctly and elegantly describe all the ultimate and intimate nature of tannage, in terms of basic fundamental principles. Leather and leather-making material science are much of rapidly evolving *arts* for that! This is not the very last model you will use to devise tannery processing strategies, as our leather field should continue to evolve with the creation of not only new, but chemically different chemical unit processes, as well as new and different mechanical means to accomplish them. In the near future drums might even disappear!

In any case, the adept tanner must always consider which are the active chemical groups in collagen/leather that react with the offered *tannery chemicals* and how to offer them at as low an astringency as required, but at the highest concentration feasible, hence low floats are

advantageous for achieving the desired rapid penetration and distribution through the cross section. Then and only then, *allow* or *change* the reaction conditions to cause higher astringency that yields higher exhaustion efficiency. In most cases, we at least know which of the collagen's reactive groups are involved, although the detailed course of the reactions that follow may be somewhat unclear yet.

Analogy between a Cross Section-Cut and an ion exchange Column

This reasoning to get adequate penetration of chemicals into collagen is in accord with concepts used successfully in dealing with chemicals involved in the use of amphoteric ion-exchange resin systems and predicting their transport characteristics through such columns. An old-time leather chemist, Prof. Dr. W. Grassmann¹⁾ had even proposed the actual use of the R_f value concept from ion-exchange resin separation technology, as a parameter applicable to deal with the penetration/astringency issues for leather chemicals coursing through the complex biological multi-charged matrix, as comparable to such an analogous *ion-exchange* column that collagen/leather would then virtually resemble. Dutta²⁾ had made parallel analyses of the problem, and refers to *resistance [to penetration]* as being due to *High Degree of Reactivity of Collagen*. The *astringency* between the chemical and collagen is inversely related to the *rate of penetration* for the chemical coursing through the cross-section. The analogy between collagen/leather acting like an amphoteric ion exchange resin in an aqueous medium, equipped mainly (but not only) with anionic carboxyl and cationic amino side-groups attached, is thus incorporated into this particular collagen/leather chemical model. The effect of pH values as well as of the chemical conditions required, such as *ionic strength*, *dielectric properties* of the medium, etc. must be considered. Built-in as well, is how to consider “masking”, both by coordination and by just simple electrostatic blockage, to achieve our objectives. Specific Hofmeister Series ions effects, that are causing the three collagen's peptide strands side-groups to be as reactive at that particular stage. Thus the manipulation of the *chemical reactivity* (astringency) of collagen's reactive side-groups, that is, the actual control over the *effective* pKa values for the amino and carboxyl side-groups of each of the individual polypeptide strands involved in conforming tropocollagen, is of paramount importance to be able to achieve tannery processing objectives.³⁾

To summarize: *high astringency* (reactivity) causes *poor penetration for a tannery chemical*, and vice- versa, *low chemical affinity aids penetration*, for those charged chemicals as mostly used in the tannery.

An array of interconnected chemical equilibriae arising out of classical biochemistry, based on work by Hofmeister, Sørensen, Levy and others, in combination with colloidal chemistry concepts give rise to a chemical model useful for the appropriate strategies for offering chemicals based on manipulating the *reactivity* of collagen. The different tautomers of an amino acid (as well as for simple peptides, polypeptides and even insoluble fibrous, complicatedly structured proteins, such as wool and collagen!) are considered by the classical *Sørensen's Formol Titration* scheme explanation.⁴⁾ These *inter-convertible*, but differing peptide structures, represent a chemical model that can be used to consider the reactivity of collagen and leather. The classical equilibriae [see chemical model *detailed later*] involved in the array have been slightly expanded by two added items: to Equilibrium **A** (left), the uncharged, “canonical” tautomer, and at **F**

(above), the water-micelle representing the triple helix. The model is able to describe how beam-house chemically purified collagen⁵⁾ reacts as it is being further processed. The non-charged canonical tautomer availability on the left side of Equilibrium **A** can be encouraged to increase, at cost to the *Zwitterion* (charged, but overall neutral!) *tautomer* fraction available on the right side of Equilibrium **A**. A *less polar* medium (including the gas phase), dehydration, higher temperatures (as it is less stable) etc. can encourage formation of the *non-charged tautomer* species. The *charged* (side-groups) *Zwitterion tautomer* on the right is more stable under *aqueous and polar* solvent conditions. There is a great *decrease* in entropy when the *Zwitterion tautomer* is formed, that is, *increased* self-organization takes place, which has been attributed to *low-entropy water polymers* interacting with the peptides involved.⁶⁾

When a *solvent fat-liquor* is used under very low float conditions, the reverse is true, and the formation of the uncharged (canonical) tautomer is then favored, thus *encouraging* an individually charged tannery chemical's penetration. The oxidation tanning process for making *Chamois* also uses carefully controlled *higher temperatures, dehydration* (salt added at a diminished float!), addition of *powdered alkalis* (discharges cationic amino groups), and the use of a *non-aqueous solvent* (cod-oil!) in order to *encourage acrolein*, an air-oxidation formed aldehyde-product, to initially react with the *uncharged amino* (–NH₂) side-group present. As in the case of *all* aldehydes, acrolein will *not react* favorably with the charged amino (–NH₃⁺) side-group such as present in the *Zwitterion* tautomer configuration. The colloidal (*water*) *aggregate* at the top of Equilibrium **F**, that represents the tropocollagen triple helix structure, has also been added to introduce to the reader the concept that the triple helix results from three polypeptide chains, mainly held together by (water-aided) H-bonds between imino and keto “backbone” elements of the different linked alpha amino-acid strands, as a consequence of peptide bond formation and resulting in increased (helicoidal originated) *hydrothermal stability*. Augmented stability additionally occurs in the case of mammalian collagen and is associated intimately with the hydroxy-bearing, hydroxyproline and other amino acid components *favoring H-bonding*, between each individual long “Jello” polypeptide strands (–R–), that are intertwined into a right-handed twisted rope-like trimer” that is the triple helix. Collagen, however, is also *additionally thermodynamically stabilized* by *electrostatic saline links*⁷⁻⁹⁾ which have key consequences on its chemical reactivity in the tannery,¹⁰⁾ as well as by *highly-ordered* water polymeric sheaths possibly physicochemical analogous in role, to the waters involved in creating surfactant-type micelles, *also* a colloidal-realm phenomenon related to the schizophrenic concurrent hydrophobic/hydrophilic character of large amphiphilic molecules, which tanners will surely need to better understand their hydration and dehydration properties.

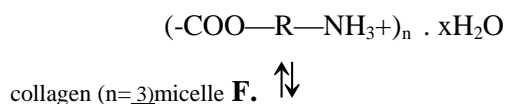
The distinction between tannage and retannage is really just only a *traditional* administrative tannery convenience, rather than a truly *relevant* chemical distinction in the fundamental chemical processing of leather, thus is irrelevant to this chemical model. The fact that shaving is traditionally done before “retannage” as not to waste any of such retanning chemicals, in shavings, greatly limits our thinking and actions as tanners regardless of actual real material cost options involved!

In the year 1931 J. A. Wilson presented a lecture titled: “*Leather, Sanitation, and Colloid Chemistry*”.¹¹⁾ It clearly describes the operational reasons for practicing tanners to mistrust leather chemists! Wilson proposed then that the concepts of colloid chemistry might yield a better outlook required of the formal chemist as a tanner for required compromises; such as to be able reconcile the *differing viewpoints* between the practical tanner and the more theoretical based leather chemist. Whether this is feasible or not, the author hopes that the practical use of this model will help at least, to diminish this philosophical approach gap mentioned by Wilson. When vats and then drums were introduced to the tanner in the early 20th century, many of the traditional recipes continued in their *fundamental design* to be still suited better for pits. Only slowly and more recently, has the drum been better employed, *at low floats and proper speed*, to its full potential as a *chemical reactor* in the tannery.

The Chemical Model

F. (above) includes the so called supramolecular water-structure ($n=3$ for tropocollagen, $n=4$ for Hemoglobin, $n=2$ for DNA); each of the intertwined (not necessarily *exactly* identical) peptides, or alpha-helix strands represented as -R-, that is then, a long “Jello” (commercial gelatin product) type strand of peptide-bonded joined alpha amino-acids with multiple amino and carboxyl side groups behaving as acids or bases, attached. These side-groups appear also distinctly in (the separated by the process of denaturation) gelatin individual-strand titration curves, whereas x^* mysteriously represents very obviously complicated chemical functions; yet to be really understood, and not just dealt as simply “waters of (organic) crystallization”, for there are several types of waters involved very differently in such structures.¹²⁾ Multiple types of complicatedly associated water is the real and effective distinction between leather and the *usual* leather substitution plastics.

The Colloidal (?) Aggregate with Water, at the top of Equilibrium **F**, is a *Micelle* when above an equivalent critical (minimum) micelle concentration (CMC) analogue value. Excessive hydrotropic (Hofmeister) *intimate* swelling of the tropocollagen triple helix ultimately brings about the irreversible denaturalization by “peptide dilution” below the CMC value, that causes the unraveling into randomly distributed coils in the solvent; that is completely separated, (“Jello”) alpha-amino acid strands, which are the building-block components of the triple helix.



Dehydration

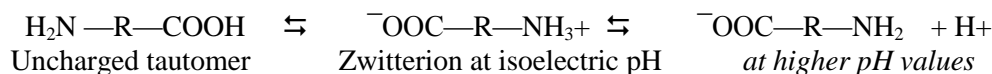
B to right as basified

← “salting-out” of solution **A**.

peptized

B.

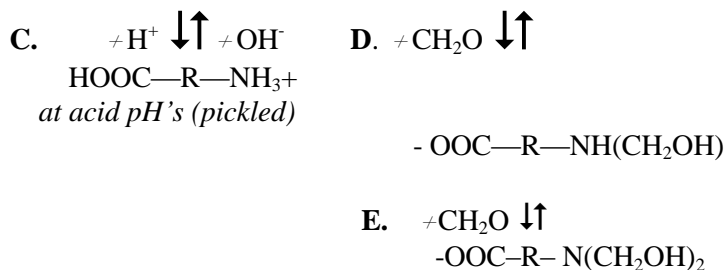
* x should obviously deal with several different types of waters, hence *interhelix* water involved in lyotropic Hofmeister effects, is more likely different from supramolecular “sheath” waters involved at other dimensional hierarchies and processes, as in hydration *stabilization of Zwitterions* associated with proton transfer and the resulting *peptization of collagen*.



“salting-in” to solution →

(neutralized)

Hydration



These equilibria (B. through E) are employed in classical biochemistry for the descriptive mechanism behind *The Sørensen Formol Titration* results, extensively observed in proteins, amino acids, and peptides; a phenomenon, by which in the mere presence of formaldehyde, causes that the value of the *pKa* for the amino group (in Equilibrium B) to drop from 2 to 5 pH units! The explanation traditionally offered, is that added aldehydes will *displace* the whole sequence of series towards D. and E., making an amino side-group proton (—NH₃⁺) in B. ***much easier to remove***, that is for the peptide species to *charge negatively*, thus making the resulting net *anionic peptide* configuration be *more reactive* towards *cationic* chemicals, and less likely to return easily back towards the Zwitterionic state. This disablement of a saline bond between cationic amino and anionic carboxyl side-groups of collagen/leather by discharging the amino has chemical processing consequences for the tanner. Aldehyde, then is effectively “*unburying*” the previously somewhat *un-reactive* carboxylic groups present in the Zwitterion tautomer, as they are themselves involved in *longer-ranged*, saline links with cationic amino side-groups in the cases of proteins, amino acids, and peptides. This *Sørensen Titration* aldehyde effect, by lowering the *observed pKa* value of the *amino-group* in the titration curves, occurs as well in even *ordinary amino acids* (-R- would be then a small group) dissolved in aqueous solutions! The fact is that the *pKa* for the *amino drops*! If there is an Debye – Hückel saline “screening” effect breaking-up Zwitterionic interactions through the solvent between even individually dissolved peptides, then the Hydration/Dehydration of the postulated Micelle in the model would act thusly on the titration curves *pKa* values as predictable from the chemical model proposed:

From B.; $K_a(-\text{NH}_3^+) = [\text{H}^+][{}^-\text{OOC}-\text{R}-\text{NH}_2] \div [\text{Zwitterion}]$

From C.; $K_a(-\text{COOH}) = [\text{H}^+][\text{Zwitterion}] \div [\text{HOO}-\text{R}-\text{NH}_3^+]$

Since the (*Ka*) acid dissociation constants for the cationic amino group from B. has the Zwitterion concentration in the **denominator**, whereas the carboxyl *Ka* from C. has the Zwitterion concentration in the **numerator**; it is to be expected that as the Zwitterion concentration changes by hydration/dehydration effects on the *peptide micelle*, the relevant *pKa* values change oppositely, and hence the display of rotation of the titration curves about the isoelectric point, this hints that the chemical model being proposed is more than just a mnemonic device to obtain the right strategies!

Saline-bond interaction between solvent-separated *Zwitterions* evidently still occurs, and is *functional*, even at the very long distances (on a chemical-molecular scale) increased up to the optimum “peptization” value, which is equivalent to about the 5 (NaCl) β e for collagen such, as was previously suggested as the likely mechanism causing the clockwise rotation of peptide titration curves.³⁾ A greater neutral salt addition causing a colloidal realm “*protein dehydration*” process occurring *in the immediate surrounding environment of the* saline links with a corresponding *decrease of the Zwitterion* fraction present as described by Equilibrium A shifting then towards the left. Even though there are alternative or additional applicable explanations for this obviously extremely complicated effect¹³⁾ the solubility decrease (salting out) of the peptide is attributed to the peptide’s dehydration[†] by the added salt. These opposite-sense changes in their observed pKa values, as per titration curves, for *either of the components of the saline bond*, just as it also occurs with more complex peptides and even fibrous proteins¹⁴⁾ as well, and can be controlled by the tanner when subjected to *varying ionic strength*. Collagen clearly displays the pKa’s clockwise rotation of acid and basic side-groups about the isoelectric point by saline increase: See, for highly interesting work done by Bowens and Kenten, *Biochem. J.*, 1948, (43)3, 358-365. Covington has the titration curves in page 22 (Ref. 20) of his book ***Tanning Chemistry***. Please see pg.361 in: <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC1274697/>

It should be mentioned that the effect on the *peptide’s amino pKa* values by the *action of aldehydes*, is in the same sense and fairly similar to the changes brought about by the increases in ionic strength, that is, its displacement towards lower pKa’s values, such as similar to *Zwitterion concentration decrease* through dehydration, but obviously by disruption of a colloidal saline bond as well! The pKa change of the *carboxylic* functions as *being nil by the peptide’s reaction with aldehydes* (which would hint then that the inductive effects in the case of simple peptides, through the hydrocarbon structure are likely minimal), but *it is shifted towards higher values* by the *increased ionic strength* induced salinity hydrating effect. That is, in an *opposite sense* to the amino group pKa displacement by the same saline hydration effect, resulting in an overall clockwise rotation about the isoelectric pH point of the titration curves, as salinity increases to the maximum ionic strength at the *optimum salting-in point* of about 5 β e NaCl salinity for tannery beam-house treated collagen. Manipulation of the *effective PKa’s values* of reactive groups (astringency) in collagen is the basis of achieving not only more efficiency in chemical usage, but for also making better leather!

These are then presumably *colloidal effects* that occur at dimensional hierarchies *greater* than the usual chemical-molecular scales of dimensions. The dissolved ordinary alpha amino acids also behave similarly to more complex peptides and proteins, as to the effects caused by Debye-Hückel ionic screening processes and by “*dehydration caused discharge of the Zwitterion*”, as inferable by their parabolic (as they salt-in and then out!) solubility curves vs. ionic strength

[†] Edwin J. Cohn in a milestone ACS Monograph #90 of the mid- 20th Century, in ***Proteins Amino Acids and Peptides***, see Ref. 4) pgs. 608-609 states: “..... Hofmeister emphasized an important element of the truth in suggesting that the phenomenon was due to “*dehydration*” of the protein by the added salt. This conception, couched in more explicit and quantitative terms, forms part of Debye’s theory of salting out. The specific influence of different salts, as enunciated by Hofmeister in his studies, are found also in the actions of salts on amino acids, indeed on the simplest gases. The striking qualitative similarity in the action of salts on the solubility of cystine and hemoglobin should be stressed. The *salting out effect* of salts on amino acids is apparent even in very dilute solutions; in proteins, likewise, its significance will prove to be even greater than earlier investigators were led to suspect. The elucidation of the nature of the effect is not a special problem confronting the colloid chemist only; it is rather a problem of extraordinary wide scope in the general theory of solutions..”

graphs. These characteristic solubility curves display effects due to salts are occurring even to (dilute) dissolved alpha amino acids, again suggesting that amino acids *Zwitterions* are still somehow significantly electro-statically associated even when present *in ordinary solutions*, in order to exhibit the aggregation and disaggregation effects reflected by their solubility characteristics under changing salinity as described by Equilibrium **A**. If the separated alpha amino-acids in solution, display these saline-bond (*peptizing*) character properties due to *Debye-Hückel shielding* by the dissolved salts; it should not be surprising then that amino acid side-group components in the triple helix individual gelatin strands, locked into the triple helix configuration at relatively close (at colloidal dimensions) separations by H-bonding, should also display the characteristics of coulomb electrostatic longer-ranged interactions occurring between anionic carboxyls and cationic amino side-groups, also likely between each of the separate strands conforming (interhelically) the same triple helix! Certainly *intrahelical* and even *intrafibrillar* saline bonds and at even higher colloidal dimensional hierarchies would still be possible, as well to also occur at even lower salinity values than the closer ranged inter-helical saline bond disruption by higher salinity values as required by the Debye-Hückel electrostatic screening at the much shorter inter-helical dimensions.

Distances between opposite charges are very important then in Debye-Hückel screening analyses. This *salting-in* colloidal effect, due to ionic-strength charge shielding action, has been called “*Peptization*” in classical colloidal terms and is paramount in determining the *chemical reactivity* (astringency) of proteins, amino acids, and peptides, towards the mainly *anionic* and *cationic* tannery used chemicals, as well as the peptide’s increased swelling/solubility *in and by water*, through the *salting-in* action at isoelectric pH values referred as “neutral schwellung” in the older colloidal literature.

These compound salt-bonds (for they also could possess possible potential H-bonds) interactions in the triple helix water-micelle, should contribute to additional hydrothermal stability to collagen.¹⁰⁾ The characteristic protein salting-in/out effects as is reflected by their solubility curves being parabolic and are not only displayed by ordinary amino acids and more complex peptides as well, but are also displayed by the surface physicochemical characteristics of an otherwise insoluble fibrous protein in just contact with water! An aqueous gelatin solution, that is: the peptide products resulting from denatured triple-helices, after the organizing H-bond structures are destroyed (by “denaturing” through interhelically Hofmeister lyotropic swelling accompanied by a *large* entropy increase); also display the rotation of the titration curves about the isoelectric point at increasing salinities, (such as Collagen does) indicating that the Zwitterionic saline interactions remain active even in the “Jello” solution, that is, after the triple-helix itself is no longer in existence itself. Remnant Equilibrium **A** features can then still be manifested in the solvent separated (randomly distributed) gelatin strand individual components of the triple helix, because of the much longer characteristic distance of effective range of saline-link interactions compared to other short-acting linkages in the molecular-chemical realm. Thus gelatin salting-in/-out phenomena is similar to those in *collagen fibrils* and even *in wool*!

When we assert that the reversal of **A**. occurs because of dehydration as being opposite to the mysteriously complicated hydration that gives rise to the Zwitterion micelle stabilization, as established by spectroscopic evidence in amino acids and simpler peptide systems,¹⁵⁾ we are

obviously speaking of dehydration effects on highly organized complex H-bonded polymeric water systems comprising the mysterious (colloidal micelle?) supramolecular water self-organizing structures at the top of **F.**, most likely involving also the saline bond's additional potential H-bonding capacity of the amine's hydrogen's, with electronegative component-elements of the supramolecular water-sheaths present in the lowered entropy (self-organizing) micelle structure. The tauntingly complex colloidal effects behind the displacement of Equilibrium **A**, need not philosophically overwhelm us into complete inaction, but we should humbly accept our obvious current ignorance, when working as tanners, as long as we can get the desired salting-in(to solution)/-out(of solution) effects and make charged reactive side-groups to discharge and recharge at our will! We should use the benefits of variable ionic strength and dielectric properties based methods affecting effective pKa's values of side-groups in processing leather, for astringency control purposes, both for enhancing penetration as well as for reaching our ultimate fixation/efficiency goals.

The Sørensen Formol titration scheme then similarly explains why formaldehyde (or better yet, work-place preferable Glyoxal, Glutaraldehyde or even Glyoxilic Acid!) in the pickle helps to chrome tan better and fix more chrome on leather (with *up to* 96% efficiency!) as so stated by the late Umberto Sammarco in the 2006 IUCLTS Congress in Istanbul.¹⁶⁾ “Unburying” anionic carboxyl's (by their effective “demi-peptization” as corresponding saline links by cationic amino side-groups are discharged and blocked by the added aldehyde) that were previously involved in said saline links with cationic amino collagen side groups, present both in inter and intra linked tropocollagen helices, and would logically cause more appropriate reactive sites for cationic metal tannage (co-polymeric-hetero-coagulation?) to be made available in collagen, at several differing dimensional hierarchical levels. Aldehydes, effectively making collagen less cationic (although not really an anionic chemical!), and chrome cationic tannins will be better exhausted.

C. (vertical) down describes the peptide species in equilibrium under acid conditions, such as when pickling (or even when **B.** is reversing to the left and then down through **C.** when an anionic re-tannage product is being fixed by the use of formic acid!). Hence this cationic protein species would be expected to be clearly less reactive to cationic metal tannins— at least as not until the actual basification is initiated! For didactic purposes only, this micelle structure in the top of step **F.** is depicted with much more structural detail as the triple helix structure, differently from those others, really also trimeric structures, but simplified in the rest of the series. Obviously all the tautomers of collagen that are already aggregated into trimeric colloidal triple-helix “micelles” (unless they were previously and irreversibly denatured into separated “Jello” strands randomly distributed throughout the solvent!) are thus not depicted as conformed as triple helices, but just for clarity sake! This seemingly outlandish affirmation that collagen, a fibrous protein, (equilibrium **F.** vertically up) as being thermodynamically analogous in a structural physicochemical sense to colloidal water-surfactant micelles, is not just by any means the author's original idea. It has variously been suggested as an insight by well-known leather scholars such as McLaughlin-Theis¹⁷⁾, Bienkiewicz¹⁸⁾ and the much respected modern chemist Vollhardt in his “Organic Chemistry” text.¹⁹⁾ The colloidal concept of supramolecular water structures (clathrates?) or aqueous-sheaths surrounding proteins at colloidal supramolecular dimensions, and is commented upon by Dr. A.D. Covington²⁰⁾ as well. The site for the relationship between Hofmeister Series hydration/dehydration ion effects occurring²¹⁾, that is

between the individual polypeptide strands associated through colloidal structured waters into tropocollagen; rather than in the free “bulk water” of the true solvent phase involved. Salting-out *in* Equilibrium **A** is probably a much different phenomenon to the Hofmeister lyotropic coagulating (de-swelling) effects occurring at the triple-helix *internal* dimensions level and always results in increased hydrothermal stability as individualized H-bonding becomes then better oriented. This will surely be a difficult, but an interesting subject to still preoccupy us. Collagen chemists need undertake many future studies, but it is perhaps too premature yet, as tanners; for we are being currently considered as ecological-political scapegoats in so many countries. We must defend ourselves here and now, through effective increased-efficiency results that need not require us yet completely delving into *long standing* unresolved controversial colloidal theories on solvation of *proteins, amino acids and peptides!*

Preliminary Trials for Increasing Chrome Tannage Efficiency

In several Latin American tanneries, chrome tannage preliminary trials were undertaken in which relatively small amounts of an aldehyde agent combined with an aluminum salt product, were added *at the pickling* stage. This caused an apparent increase of *chrome utilization efficiency* compared to the usual conventional processes employed in that tannery. **Appendix A**, at the end of this paper, shows the results.

Is chemical linkage involved in Tannery Processing? What Types of Cross-linking?

Many tanners and leather chemists are very predisposed to consider the concept of *cross-linking* by chemical means as being at the heart of all tannery processes. However, they prefer to accept high-energy *covalent* cross-linking as the really the “politically correct” explanation, (as compared to a weaker, but much longer-ranged *saline* links), although naively paying some attention to hydrogen bonding concepts as being very “modern” and thus somehow ought to be included in any discussions of real current scientific value!

Several Relevant Chemical Structural Linkages that *can* apply to Collagen/Leather

It is useful at this point to make some very succinct and elemental remarks on the properties of various applicable chemical links as that will help guide us on how to understand the workings behind this model being presented for operating under actual tannery situations. Of all the chemical links, the salt-bonds primarily caused by electrostatic (coulomb) forces, are the longest-ranged and thus are the *more relevant* in the regimentation of supramolecular (nanometer-colloidal) dimension processes as opposed to smaller yet, chemical-molecular scales of dimensions, where *some* covalent bonds can be involved (such as with aldehydes) in the tannage of collagen. The *inverse square force* attractions (or repulsions for like charges) are generally the first step involved in most tannery chemical processing, because of their longer range. In dyeing, for example, where anionic dyestuffs are attracted, through their anionic sulphonic groups, to the cationic amino groups of collagen/leather at typical colloidal-range of dimensions, but generally only later, are the dyes affected by other, closer-ranged, more usual chemical-bond forces such as covalence, coordination, hydrophobic attractions, Van-der-Waals attractions, H-bonding, etc. etc. that are typically ruled by various -6 through -12 *exponential force laws* acting only *at much*

shorter distances than does the coulomb inverse square (-2) exponential law. These later types of chemical *short-ranged* forces are generally the ones involved in the *final* fixing of a dyestuff, as the leather is drying and the distances between reactive groups shorten to those then involved in typically chemical, rather than colloidal dimension events. One of the characteristic consequences of these electrostatic inverse square forces, is that collagen's saline links can be broken-down by starting to decouple saline bonds, at first at the largest dimensional ranges between layers of fibers, by the shielding attenuation on charge interactions caused by ever increasing concentrations of dissolved ions (such as those resulting from *mid-series*, relatively non-swelling/non-coagulating, Hofmeister ions such as resulting from KCl, or NaCl), and as described by the *Debye-Hückel theory of solutions*. Dr. Covington refers to this colloidal-dimension phenomenon as the "*smearing-out of charges*" when referring to the presence in collagen of salt-like links.²¹⁾ Saline links in collagen can then occur (as well as decouple!) at different hierarchical dimensional levels depending on *ionic strength* (and of course pH!) of the of the gross aqueous medium (bulk solvent) surrounding the proteins. Thus ionic strength control becomes a very important factor in seeking the somewhat mysterious colloidal state of maximum "peptization" (at relatively lower ionic strengths, or "salting-in") by breaking apart by "*ionic screening*" the longest-ranged saline links first, with a resulting increase of available reactive (water-dipole separated, that *is swollen*) charged sites available to tannery chemicals, but the basic tropocollagen structure, although maybe somewhat weakened by this gentle distortion from its fully helicoidally most-stable geometry, remains essentially intact. Thus saline links can be uncoupled by the electrostatic screening effect on the opposite charges between the intra and inter strand bridging bonding sites on collagen and the appropriate charged amino or carboxyl collagen side-groups present. In this case, it is the ionic strength as a property of *the bulk solvent* that is important. The primarily electrostatic saline-link can also essentially vanish through use of stronger yet saline medium, as a *strong* colloidal level dehydration eliminates the very formation of Zwitterionic individually charged groups ("salting-out"). In this last case, uncharged tautomer canonical configuration at the left in Equilibrium **A** more favorably occurs; the resulting bio-structures will also loose the additionally stabilizing saline bonds. Though these two phenomena appear closely inter-related, as salts can be involved in both cases, the nature of *peptization* breaking-apart of saline bonds through charge shielding by ionic strength of the *bulk solution*, and thus results in *enhanced reactivity properties of collagen* towards charged chemicals; is completely different from the saline bond absolute disappearance because of the severe dehydration caused discharge, as a proton re-migrates from amino to carboxyl, and thus minimizing the proportion of Zwitterion tautomers present. This effect is not due to the *bulk solution solvent* ionic strength properties, but by *specific effects of lyotropic de-swelling Hofmeister chemicals* affecting the *waters in between the individual tropocollagen alpha amino strands*, that is, at the micelle-colloidal level of dimensions. As a matter of fact, solvent dehydration can cause Equilibrium **A** to reverse to the left *without any salt even* being present! The different natured salting-in and salting-out colloidal effects are inherently, but somewhat simplistically (elegantly?), described by equilibrium **A**. The first (toward the right) involves just a little salt added, whereas the second (towards the left) involves lots! It must be further emphasized that the placement inside the triple helix of either of Hofmeister Ion Series (at either extremes of lyotropic swelling or de-swelling character) should affect not only the state of internal hydration of triple helix specifically¹⁸⁾, but in addition these ions could also contribute to the *gross solution's bulk ionic strength* electrostatic shielding (at specific larger distances) by the

Debye-Hückel *salting-into solution* effect. This should hint at the exasperating difficulties involved in stating quantitative expressions describing the differences between these two types of colloidal events, and perhaps both could be occurring concurrently *at different* dimensions, but at the same overall salinity! At really higher concentrations of salinity (about 15 β e NaCl and higher!), the electrostatic shielding begins to affect links spanning shorter distances still, but eventually the strong dehydration of the Zwitterion H-bonded structure conforming the supramolecular water sheaths, but not the bulk solvent, causes the saline links to vanish substantially in collagen, as it turns into pseudo-leather! The skill required of a tanner is how to use salt-addition wisely by the use of appropriate float size.

Saline-links do also affect many of the Macro-properties of Leather

The “*hardening*” of leather/collagen upon drying is related to compound saline links forming between *fibers* and *layers* of fibers at higher hierarchical dimensions. Water can act as a “fiber lubricant” by dissolving salt bonds, thus separating charges while water is present in excess, but water removal by *severe dehydration*, such as by successive acetone washes, then bars the saline link formation for lack of *Zwitterions present*, as the relevant Equilibrium **A** is substantially reversed towards the left by such an extensive solvent dehydration method, and collagen then becomes *pseudo-leather*. This is the theoretical interpretation of Dr. Azdet’s visionary ecological processing project proposal that might even eliminate (re)tanning drums.²²⁾ Pseudo-Leather dries soft (but remains very avid for water) rather than the hard, horn-like, but not easily wet-able, parchment. An anionic fat-liquor fixing on the charged amino-end of the saline link, disables it from re-coupling by electro-statically “masking it”, and thus avoids the reformation of the extended hardening saline-links between structures, while the hydrophobic, non-polar, “other end” of the oil chain is said also to “lubricate the fibers” as the non-polar, solvent-like organic character of the oil also does deter electrochemical saline bonds from effectively reforming because of the lack of Zwitterion charged configuration available, needing a polar aqueous medium present to form. In other words, a non-polar, *organic*, “other-end”, of a fat-liquor chain can similarly solvent-dehydrate collagen thus *appearing* to “lubricate” fibers! Aldehydes by blocking-off uncharged amino groups and preventing them from recharging, then tend to give softer leather because of the destabilizing of the “hardening” long ranged, saline links. Aldehyde tannage, in general, also tends to loosen the grain-break as well and can be very useful for achieving dramatic grain-milling effects sought to camouflage defects, in lieu of leather-hardening (printed) embossing. Thus fat liquors provide “*half*” of the required effect for turning collagen/leather towards the *pseudo-leather*, defined by: “as lacking too many extended saline links in the polar aqueous medium present! Actual “*Leathering*” thus involves some “dehydration”. Deep application of waterproofing hot-wax applied in the finishing, affects greatly the *temper* of the resulting leather by decreasing the Zwitterion tautomers present as the hot wax displaces water in the substrate. The Zwitterion conformers are the ones ultimately causing extended, leather hardening, salt-links. Sometimes for these reasons, a resulting loose-grained crust leather can be substantially salvaged by a slightly acid (pH 4.5-5) water and formic acid wash or spray, followed by careful mulling and re-drying, because it causes some saline-links to reform as some previously discharged amino groups are made cationic again, and tend to re-couple with relatively more distant still existing anionic carboxyl sites, thus strengthening of the multiple very long range, but weaker saline links between the grain and corium layers that can

often critically determine the “break” character of leather. This “first aid” treatment, however, does tend to harden (that’s why it works at all!) the crust a little upon drying. This, however, can save the day for a harried tanner because of a VIP customer wanting *his* particular leather’s break repaired by “a supposedly competent” and adept tanner that “should know what he is doing”, rather than solving the defective break problem by just conveniently selling-off the misbehaving leather to a potential competitor, at a discount to boot! This type of “art” is what makes a tanner into “a real scientist” in the customer’s eyes! Any mischievous boy that has wet his new shoes by purposely walking through a slightly acid (CO₂ containing) rain-water puddle, is aware of this particular *advanced grain-tightening* technique, especially if he dries-out his leather shoes very quickly, before his parents come home, and keeps quiet about resulting blisters caused by having to “break-in” his shoes all over again!

The purification of salted collagen from presumably electrostatic-attached impurities such as proteoglycans, décorin, etc., is performed usually by a 10 β e common salt (10% b.w.) solution in early soaking (~5 hours). As collagen starts to lightly de-swell at slightly higher salinities after saline bonds to these impurities have been decoupled by slightly higher ionic strength Debye-Hückel shielding of the linking charges, and thus long-chained, previously saline-linked impurities can then be easier washed-out then at the 10 β e salinity level when collagen is also slightly additionally de-swollen from the fully neutral “maximum salt swollen-in” (peptized) state, occurring at approximately 5 β e salinity. Then, as the ionic strength of the medium increases after early preliminary rinse-washes, we get at first the decoupling of the longer ranged saline-links of the actual already purified collagen components (rather than *of* the un-removed impurities) as between fibers, in differing layers, and even perhaps some Debye-Hückel shielding decoupling between still at some more distant interacting fibers, in specific layers. The resulting uncoupled, but still charged amino and carboxyl groups, remain *protruding out into the solvent water* (thus making the peptide *more soluble* and tanning reactive!), but much too electrostatically shielded from their further-away opposite charged former “targets”, to be able to reform the longer-ranged intracollagenic saline-links at the highest of leather’s hierarchical dimensions. Collagen then becomes more chemically reactive towards anionic or cationic chemicals in its optimum “salted-in” or “peptized” condition.

Thus the “peptized configuration” version of fibrous collagen is a direct consequence of Debye-Hückel shielding of electrostatic attractions comprising saline links being uncoupled by bulk-solvent action at lower ionic strength regimes. Then, at higher ionic strengths still, such as occur during the dehydrating pickle at early tannage stages, some compound saline links between triple helixes themselves might be completely split-apart, but many in this case just simply vanish because the stronger dehydration ends the possibility of saline link existence because of substantial reversal of the Zwitterion formation Equilibrium **A**, that actually eliminates most opposite charges as canonical tautomer is favorably formed!

Dehydration many times will actually *increase hydrothermal* stability as *interhelically* H-bonding reorients their action. This type of Zwitterion discharging should also affect the ease of diffusion of charged tannins through the collagen fibrous charged bio-maze. Nevertheless at these higher ionic strengths some closer ranged intra tropocollagen saline links would be expected (at least intuitively!) to decouple, but detailed Debye-Hückel effective distance-of-

shielding calculations based on tropocollagen intimate dimensions, need to be carefully made with the collagen's structural knowledge now available.¹²⁾ Some charged amino side-groups can remain, as carboxylic groups are immediately blocked and discharged by the excess of protons at pickle pH's. The remaining few cationic amino groupings from the fewer Zwitterions left under the extreme saline dehydrating conditions, are "masked" (that is blocked) by the excess anionic sulfate ions present, because sulfate as a Hofmeister (dehydrating at the tropocollagen internal dimensions) coagulating divalent anion that would bridge electro-statically, cationic amino groups attached to the triple helix structure, forming saline bonds and even providing *some additional hydrothermal protective stability* to the delicate collagen structure under such harsh acid pickle conditions. These arguments are not being given to negate, in order to just be able to ignore, the occurrence of severe osmotic swelling by like-charge repulsions at somewhat higher dimensional hierarchies (at a *virtual membrane* level?), caused by acids and bases as *Donnan Forces* are also easily controllable by salt presence as well; but rather to place specific Hofmeister lyotropic swelling/deswelling effects of the triple helix itself into a proper dimensional hierarchy perspective! This is why *basic chromium sulfate*-containing (Hofmeister coagulating and dehydrating $\text{SO}_4^{=}$) tanned collagen yields "linked-and-locked" by saline-links, superior tanned leather (Ts~115°C), whereas *ordinary chromium chloride* (without the extra linking sulfate amino-bridging) treated collagen yields *rather ordinary* Cr tanned leather (Ts~85°C) that is "locked" but not "linked"!

Paying attention to fur processing can be illustrative as to how collagen behaves to hydration/dehydration phenomena under approximately neutral conditions, as the lack of required mechanical removal of keratin skin components in early processing of fur,²³ without alkali present, yields obvious and much simplified clear-cut neutral swelling interpretation of the soaking processes, although the key (limiting/enabling) salinity values are probably somewhat different to those affecting the usual purified collagen, that has gone through the usual alkaline beam-house processing. Initial neutral fur soaking under increasing saline conditions yields a maximum swelling, as determined by weight, at about 4.8 βe salinity. Presumably this salting-in colloidal process yields some multi-charged "peptized" zones where the solvent water interacts by separating charges due to salting-in and is taken-up by the electrostatic charged pelt, as Equilibrium A is displaced towards its Zwitterion configuration that had initially caused charges to form. As ionic strength then exceeds equivalent salinity values given by about the 10-11 βe zone, the upper limits for some of the residual neutral swelling to be still present, and then at to de-swell because of salt-caused higher salinity still, collagen begins dehydration (affecting A.) towards and past the base weight obtained in salt-free water under no-salt added, equilibrated, minimal peptization conditions. Eventually because of increased *dehydration* and discharge of the *previously peptized* side-groups that had interacted with the solvent, actually then collagen de-swells below the base intermediate weight, to display negative (more dehydrated still) weight values (at the right side of the maximum swelling peak occurring at about 4.8 βe), at about 15 βe and higher salinities. This implies that Equilibrium A then starts to reverse towards the left because of the *colloidal* "salting-out" dehydrating action on collagen's *water-stabilized Zwitterion* configuration and thus becomes less reactive still to the water as the solvent and, of course, less astringent to any charged chemicals being offered, aiding their penetration through the *less polarized* amphiphilic bio-matrix maze. Hence for better penetration of *any charged chemical*, low-float with a relatively *high gross ionic strength* (15 βe plus NaCl equivalent) is

recommended. Heidemann gives this recommendation for accelerated penetration for highly cationic multinuclear aluminum²⁴⁾ tannins that are not as readily “sulphate-maskable” into anionic species such as regular chrome sulfate readily *is* at pickle pH’s, under higher sulfate concentrations! Similarly, since aldehydes *react only with discharged amino* side-groups, higher salinity makes the non-ionic aldehyde tannins more astringent towards collagen, although aldehydes are traditionally and widely considered by *most tanners* as being *anionic* chemicals! This idea probably results because of the blocking of the uncharged amino groups by aldehydes towards the uptake protons, is thus limiting the maximum cationicity possible in aldehyde-treated collagen/leather. This gives an erroneous impression of inherent anionicity contributed to collagen by aldehydes. Thus collagen/leather, under stronger, dehydrating, saline conditions, becomes a much better avid scavenger of *unfixed aldehydes*, than that as with no salt present, pH conditions being equal! This would be a case where a little extra salinity at very low floats would help greatly to eliminate unfixed (free) aldehyde discharges in wet-white processing into the effluents; –which case, some salt or excess free-aldehyde presence– would be considered ecologically worse? Thus salinity control should be employed as well to better exhaust highly toxic aldehydes employed in toxic metal-free tannage. Thus reversing peptization (A. to left) fixes more uncharged aldehydes as its action is chemically very different from most ordinary anionic/cationic tannery chemicals. The often heard, and reckless requested “*suggestion*” made by not too well informed tannery administrators (some times as the owners!), to diminish *all salt* offers drastically, in all processing recipes, in order to attempt to obtain ecological good-will and acceptance by the chemically *not-too-sophisticated* pollution control authorities, by this action; this could possibly cause much *more harmful* pollution (as well as possible destructive osmotic swelling in pickling!) due to wasted offers of more environmental sensitive chemicals of higher toxicity, than really common salt should really be considered, because of the decrease in collagen’s specific reactivity, due to the lack of salt-induced discharged amino groups extent! By the very use of very low floats in good drums, with the possible use of a grain-protective slipping agent, *if so required*, the salinity levels needed for the required decoupling of saline links, as well as if needed to diminish the Zwitterion tautomer fraction for penetration enhancement purposes, can be obtained without ever over-loading the combined global effluents with the salts required at higher conventional usage floats. The intentional use of Glauber salt in dyeing processes, such as wool and fur dyers customarily do, would at least produce a salt-anion greatly precipitated by lime (from the usual associated beam-house waste) instead of the much too difficult to remove chloride anion! The hydration of *peptides, amino acids and proteins* under the proper saline ionic strength *encourages* the Zwitterion formation and gives rise to the *chemically charged (peptized) side groups*, the ones mainly reactive to most of the usual tannery chemicals, such as dyes, etc.

There are exceptionally homogeneous resulting and level-dyeing nubuck recipes (of the two-step kind) that obtain very rapid penetration of the anionic dye-stuff through the crust, in a very small, cool float, by the joint addition of commercial chrome tanning salt powder (about 1/3-2/3 of the dye-weight offer) and the *powder dye-stuff* (which traditionally contains substantial amounts of Glauber salt as well!). Since the chrome itself is greatly anionic under the excess of sodium sulfate present (up to nearly 50% b.w. in many commercial chrome tanning products!) and since the sulfate anion, a specific Hofmeister Series coagulating (tropocollagen dehydrating) divalent anion, because it associates by salt-linking two cationic amino groups (of which there

will be few anyway at the usual higher initial dyeing pH conditions, as being favorable initial crust dyeing penetration conditions) would fit-in in the chemical model towards the right-side of Equilibrium **B**. The weakly reactive (partially dehydrated at higher ionic strength because of low float and lots of sulfate acting on the collagen triple helix) as Equilibrium **A** reverses towards the left to yield more of the non-astringent discharged canonical tautomer; after the desired dye penetration is quickly achieved under these relatively un-reactive conditions. Following with a *hot, large* (250-300%), *formic acid containing* bath added to help fix just about all of the dye-stuff, by dilution, such as to produce low ionic strength conditions and switching the Equilibrium **A** back towards the relative mayor abundance of the Zwitterion tautomer, of much higher astringency towards an anionic dye. As the chrome complexes themselves become more cationic with time, temperature and the violent drop in sulfate-concentration caused salinity; the *dyestuff-colloidal anion-aggregates*, also *disassociate* from their own “surfactant micelle-like” aggregated-conditions, causing *increased dye* astringency as well. Many sulphonic-equipped dyestuffs often display amphiphilic colloidal-aggregation micelle behavior similar to surfactants as well! Chrome then contributes ever so gradually by its own astringency enhancement factor, as it rejects protons from re-fixing on ionized carboxylic groups by their chrome tannage, which then causes still more dye-astringent charged amino groups to occur. Ordinary Chrome behaves thus; as a dual-action auxiliary (dye-astringency leveling initial anionic action followed later as a dye-shade enhancer!). Thus basic chrome sulphate tanning salt is in effect, a self-regulating dyeing auxiliary for best dyeing purposes, *if used properly without pre-dissolution to delay its shifting to cationicity*, to control desired course of charge changes. The considerable additional benefit of re-chroming the leather also helps the quality of the dyeing operation as well! These examples also demonstrates how to operationally use this proposed chemical model, even if you do not wish to believe in colloidal science applications in the field of tanning, because of previously held convictions.

In Dr. Jaume Cot’s highly sophisticated collagen model presented at the joint ALCA-IULTCS congress in 2007²⁵⁾; he clearly and emphatically states his opinion that the chemical forces affecting fibrils and the *higher hierarchies of dimensions* of collagen such as fibers, *are electrostatic in nature rather than covalent*. Thus the compound saline links between collagen carboxyls and cationic amino groups consist of an electrostatic bond probably augmented energetically as it is compounded by up to three (most likely, just two) possible H-bonds between the amino-hydrogen and electronegative component elements that may or may not be present in the immediate neighborhood! But if there is a *colloidal supramolecular* water-sheath between the protein triple helix fibril-structures, different from the Hofmeister Series sensitive inter-tropocollagen water; it can be involved with the amino-hydrogen through H-bonds as well, and electro-statically with carboxylic elements of saline bonds attached through and between the more hydrophobic remnants of the triple helix, that is, in-between the polar side-chains and the hydrophilic zones due to the more polar back-bone proper. The *Zwitterion formation* clearly involves intrinsically the transfer of a proton through a water “wire” acting as a conductor! The title of a suggestive paper is presented such that it might entice some into delving into more details:

“Thermodynamic Description of a Contact and Solvent-Separated Ion Pair as a Function of Solvation: A Model for Salt Bridges and Proton-Transfer Reactions in Biology”

This paper by Beeson and Dix²⁶⁾ clearly stresses the importance of solvent polarity in not only affecting proton transfer in protein configurations, namely Equilibrium **A**, but deals with H-bonding energetics in peptide stabilization as well. The question of *Zwitterion* formation and energetic, steric and thermodynamic factors, that then clearly affect the shifting of equilibria (such as **A**), are discussed in the M.Sc. degree thesis of Mr. Erik Tung-Lam Cheung²⁷⁾ that makes special mention of the significance of the proton transfer reaction needed for understanding the *Zwitterion tautomer formation*, that is often just ignored by tanners. These last theoretical background considerations are being then presented for those interested in delving a little deeper into the topics of this presentation stressing *importance of saline links* as being vital in augmenting of the overall thermodynamic stability of the triple helix micelle and thus as an insight into the basis of the proposed concept by the author and referred to as “thermodynamic originated”³⁾ *steric hindrance* as being the cause for the various possible reasons for unreactive (“buried”, “unavailable”, “self-masked”, etc.) carboxylic and amino side-groups behavior as reported in the biochemistry²⁸⁾ and tanning literature²⁹⁾. Many more reactive groups[‡] need to be made available by *ionic strength control*, saline-bond break-up, etc. in tannery reactions to help approach a more complete stoichiometric outcome, to not only avoid chemical waste, but to actually make *better* leather.

The Thermodynamic Stabilizing Saline-bridging (*) within the Various Hierarchical Dimensional Structures of Collagen can be Described: Collagen — R'-COO⁻ * + H₃N-R''—Collagen.

The basic principle concerning enhancing reactivity of collagen to most tannery chemicals, is hereby being presented to the tanner: any offered chemical (including **H⁺** and **OH⁻**!) that decouples this saline link, *very widely* present *throughout collagen at several* different hierarchical dimension levels, by chemically fixing on either the anionic carboxyl or cationic amino, then frees gradually the remaining component of the electrostatic salt-link, denoted by the asterisk *, to react with an opposite charged tannery chemical being, or to be soon, offered. Thus the well-known beneficial action of a commercial syntan-masked chrome tanning products³⁰⁾ is due to the anionic sulphonic aromatic syntan component fixing on to the cationic amino, as it then frees then the corresponding anionic carboxyl of collagen, to react with any cationic chrome present, as it rapidly turns into cationic tanning complexes from the supposedly *freshly-dissolved* highly masked sulphonic anionic complexes formed. Sulphonic syntans can be also considered to be as specific Hofmeister Series coagulating (tropocollagen dehydrating) *weakly* tanning anions. Even when so called masked-chrome syntan preparations are added in the presence of ordinary basic chrome tanning sulfate salt, an increased *total chrome* absorption by collagen results and the produced leather itself is much superior to that obtainable by adding only ordinary commercial chrome alone. This is often being reputed as being because of the “combined” action of the dual tannage concurrent processes occurring, that it is both a sulphonic aromatic syntan *anionic tannage* and that of *cationic chrome tannage* itself. The beneficial aspects of the effect are also often attributed as well, to the “masking effect” of the *syntan on the chrome*, but it

[‡] A.D. Covington²⁹⁾ makes the surprising calculation that only about one in six collagen carboxyls available actually reacts with chrome tannins offered. A *small increase in reactivity* should then remarkably improve efficiency!

should be made perfectly clear that in cationic aluminum tannage as well, is improved by the concurrent addition of such an anionic sulphonic syntan, even though in the case of aluminum, the much weaker metal-masking is occurring, because of its lesser coordination capacity, is probably then not too much of a real factor at all! Certainly the co-use of aldehydes to effectively disable the aforementioned saline links through more permanent discharge of cationic amino groupings, also exhibits better chrome uptake in tannage as proposed by Umberto Sammarco and others, and is also due to the synergistic effects of decoupling the saline link primarily, as described by the previously discussed Sørensen Formol titration effects on chrome fixation, and in that particular case, there is certainly *no additional coordination chemistry advantage* involved! Certainly the remarkable increased chrome efficiency effects by offering during pickle Glyoxilic acid (HOOC-CHO, Hoechst's Feliderm CSTM) are explainable as the aldehyde function blocking amino-side groups against salt-links with carboxyls, thus also disabling saline links.

There is a favorable dye-leveling effect involved by the blocking action by aldehydes of charged amino formation, especially those in scar tissue! Scar tissue being somewhat of nature's emergency "rush-job" for repairing disrupted normal collagen tissues, even if temporarily, perhaps then has less cationic amino groups favorably placed such as to be properly involved in the additionally stabilizing saline-links, and thus scars will naturally dye more intensely because of having more *uncoupled cationic amino* groupings than, the slowly-growing, more stable, more perfect, highly helical character, "triple A rated" collagen! The combined use in the pickle of aldehyde and a cationic Hofmeister lyotropic swelling agent (such as an aluminum salt!) that lowers the hydrothermal shrinkage temperature of pickled collagen by about 20-25°C, and has certainly been shown to improve chrome uptake and results later in more intense dyeing as well (see appendix A). If a neutralized naphthalene sulphonic syntan product is part of the subsequent basification as well, fuller chrome-use efficiencies perhaps even surpassing the Sammarco's 95+ percent fixation are possibly achievable, even without reaching the recommended basification temperatures for better chrome exhaustion at 48-50°C, because of the *traditional lack* of adequately powered drumming equipment *at most* tanneries! The fact is that gentle and controlled destabilization of the triple helix itself by mechanical *energy inputs* such as described by the Hinsch *mechanical drumming effect*³¹⁾ analysis, *ultrasound energy* input, *generalized* temperature increase, *lyotropic swelling* agent pretreatments by Hofmeister Series swelling ions (or similar acting chemicals, such as *urea*!) that gently destabilize thermodynamically the triple helix by the straining of the helicoidal structure (to more resemble and act as scar-tissue does!) as it lyotropically swells and some saline links probably become undone, and are presumed to make collagen more chemically reactive. Ionic-strength induced "*peptization*" by Debye-Hückel extensive shielding of the saline bond charged components, also has in common, an observed associated increased chrome tannins uptake upon the basification dilution, which would seem congruent, at least, with a partial additional decoupling of the saline bonds being hereby proposed as an explanation for all these well-known effects,³⁾ and thus allow us a closer approach to the achievement of *nearer stoichiometric* goals in the tannage reaction (regardless whatever it's "true nature" *might be*!) to yield better leather and waste less chrome. Thus overcoming *thermodynamic-stability originated steric hindrance to chrome-tannage* by decoupling additional collagen stabilization by saline links (such as heat-denaturing also does!) will increase the availability of reactive carboxylic groups on collagen. This tried and seemingly successful

improvement philosophy needs to be practically implemented to effectively demonstrate to the pollution control authorities, that we, as tanners, are doing something tangibly real about unnecessary chromium pollution, even at the commercial risk of having to give-up some possibly now fashionably-current sales of metal-free leather!

One lyotropic Hofmeister cationic tropocollagen-swelling exemplary agent⁸⁾ often employed in these high efficiency chrome tannages is a small amount of a soluble aluminum salt in the pickle! This will actually lower the Ts by about 20° C³¹⁾. There has been extensive discussion of the benefits of combining chrome tannage with an aluminum pre-tannage³²⁾ to better utilize chrome. Covington³³⁾ refers to it as a “*catalytic*” effect of aluminum on the chrome tannage reaction, resulting in the development of the aluminum-containing ChromesaverTM family type of tannery chemicals, but not with much actual usage for shoe upper leather manufacture occurring because of the tanner’s reluctance to change from the accepted practice for a mixed mineral tannage; such as with aluminum, when used as a substantial component of (re-)tannage, requires a much higher end-pH of basification than that of chrome alone.³⁴⁾ This fear that ending basification at higher pH (5.8-6.0), is considered most likely, by most tanners, as being a sure way for causing loose grain, and unfortunately, as well, the tanner’s perception that one should not expend even inexpensive and relatively ecologically perceived-as-harmless chemicals such as inorganic aluminum, in the making of shavings. This causes tanners to mistrust these recipes from the very beginning, although *they are known* to appreciably increase chrome-tanning efficiency. There appears that no objection is found in using excessive and fairly toxic aldehyde combinations in some wet-white-type production, in the interest to yield all-metal-free leather!

The colloidal concept of “peptization” (salting-in) of collagen caused by mild salinity, as well as the decrease of astringency by the use of much higher ionic strengths (salting-out) have all but disappeared after WWII from most tanner’s repertoires of conceptual tools. Probably because of the difficulties in clearly understanding the application of these colloidal concepts, but the mastery of these, is essential to the use of this chemical model being proposed (especially in the application of manipulating Equilibrium A) for the intelligent offering of chemicals to collagen/leather; hopefully this presentation will help the tanner remedy this situation. Perhaps more enlightening research will soon arise to help explain these somewhat mysterious appearing and mistrusted colloidal processes better to the tanner.

As technology evolved from the use of pits to that of drums, where ionic strength control, mechanical effect control, temperature control, float size control, etc. are more feasible. Thus a drum should be as thought of being more of a chemical reactor, instead of just an “improved” motorized pit. Unfortunately most processing recipes remained still heavily influenced by classical pit-technology-thinking, and need to be better adapted to more modern conditions. It is hoped by the author that the use of this model based on “expected astringency” between collagen and most common chemicals will so help.

Appendix A

Estimated Improvement in Chrome Uptake Efficiency:

Normal Standard Tannage 6.5% BCS offer.

Average according to SENAI, nine samples= **5.18%** Cr₂O₃ contents

ABC Leder reduced chrome tannage 5.0 % BCS offer

Average according to SENAI, nine samples= **4.68%** Cr₂O₃ contents

Difference in ratio of Chrome offer: 6.5%/5% = **1.30**

Difference in ratio of results: 5.18%/4.68% = **1.11**

Estimated increase in efficiency by addition of 0.3% of Al₂(SO₄)₃ and 0.5% aldehyde tanning agent at 40%, to pickle at *reduced* chrome offer tannage: ~19%

Full-production tannage trials with about 10 metric tons of un-split limed hides were performed. Chrome stratigraphic contents performed on wet-blue by the Brazilian Leather Institute, SENAI. The data obtained seemed to have little dispersion and standard deviation information. No attempt was made, to assess the individual independent contributions of the aldehyde, aluminum and other possible contributing factors, to the increased chrome tanning efficiency, but just to show that there is probably still room to clean-up our act as tanners!

Acknowledgements

- I wish to thank my clients past, J.H. Lowenstein Inc; and present, for allowing me to try in the field somewhat unconventional formulations, especially, Moyle Mink and ABC Leder Grupo Andino S.A. (CEO I.Q. Héctor Mario Agudelo) that is undertaking these projects of minimizing tannery effluents through increased efficiency in chemical usage. I thank ALCA for their recommendation for this presentation and the people that provided helpful discussions and are just too numerous to list!
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