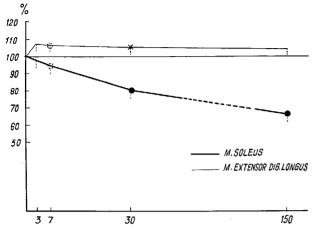
values for extensor muscles, which led to a levelling out of the flexor-extensor difference. These results, together with observations described above, are considered as evidence for the increase of excitability of motoneurones in the anterior spinal horns for extensor muscles after de-afferentation.



Weight changes in m. soleus and m. extensor digitorum longus after section of the dorsal roots  $L_1$ - $S_1$  proximal to the spinal ganglia in the rat. The results are expressed in percentage of control muscle weight of the contralateral extremity ( $\gamma$  axis). Muscles were weighed 3 days (7 animals), 7 days (26 animals), 30 days (32 animals) and 150 days (5 animals) after the operation (x axis). Values marked with a cross are statistically significant for p=0.05, with a ring for p=0.01 and with a dot for p=0.001.

These changes in excitability lead to muscle atrophy in extensor muscles after de-afferentation. We studied weight changes in m. soleus (physiological extensor, antigravity group) and m. extensor digitorum longus (flexor group) after de-afferentation in cats and rats. It was found that m. soleus progressively atrophies after de-afferentation, while no weight changes were noted in m. extensor digitorum longus (see Fig.). Similar results, only less marked, were obtained on m. gastrocnemius and m. tibialis anterior.

The differences due to changes in excitability are further shown in experiments in which muscle hypertrophy or recovery of weight was evoked. On the one hand, we used unilateral amputation and daily "exercise" of the remaining de-afferented extremity by running rats (13 animals) in a rotating drum as a means of provoking muscle hypertrophy. On the other hand, reinnervation of de-afferented muscles 10, 20, and 30 days after crushing the nerve in 45 rats served as a model for weight recovery after denervation. In both cases the results were in agreement with our previous findings. In the amputation experiments, it was found that m. soleus atrophies, while m. extensor digitorum longus hypertrophies significantly more than in control animals. Weight recovery during reinnervation is significantly slower in m. soleus and a little faster in m. extensor digitorum longus than in control muscles.

These experiments also show that the atrophy of m. soleus after de-afferentation is not so much due to the loss of adaptational ability of de-afferented muscles to increased work exertion, as to the increase of excitability in extensor motoneurones in the anterior spinal horns following de-afferentation. De-afferentation atrophy of extensor muscles of the cat and rat may be considered to be a peripheral metabolic consequence of this increased

excitability of extensor motoneurones. We do not agree with Kuré<sup>11</sup>, Wyburn<sup>12</sup> and others, according to whom dorsal roots contain trophic nerve fibres, because a similar course of muscle atrophy was noted in extensor muscles after spinal gangliectomy in the rat as after section of the dorsal roots proximal to the spinal ganglia.

We may conclude that metabolic recovery (trophic) processes are influenced by the functional state of nerve centres, and that inactivity *per se* cannot fully explain the onset and rate of muscle atrophy.

P. Hník

Physiological Institute, Czechoslovak Academy of Sciences, Prague, February 21, 1956.

#### Zusammentassung

- 1. Die Deafferenzierung der hinteren Extremität bei Ratten und Katzen verursacht eine Neigung zur Extension, welche sich 7–10 Tage nach der Operation zu entwickeln beginnt; diese sehr ausgeprägte Neigung wird durch eine erhöhte Erregbarkeit der motorischen Vorderhornzellen erklärt, welche die Extensoren innervieren.
- 2. An der deafferenzierten Extremität atrophieren die Antigravitationsmuskeln (m. soleus und m. gastrocnemius), während man bei den Flexoren (m. extensor digitorum longus und m. tibialis anterior) kein Anzeichen von Atrophie findet.
- 3. Die Deafferenzierungs-Atrophie der Antigravitationsmuskeln ist als eine in der Peripherie eintretende metabolische Folge des veränderten Funktionszustandes der Nervenzentren aufzufassen. Diese Ansicht konnte mit Hilfe weiterer Experimente (Reinnervation und Arbeitshypertrophie) bestätigt werden.

 $^{11}$  K. Kuré, Y. Nitta, H. Matsuura, and M. Tsuji, Z. ges. exper. Med.  $60,\,250$  (1928).

<sup>12</sup> R. Wyburn-Mason, Trophic Nerves (London, 1950).

## On the Stability and the Reactions of the H<sub>2</sub>O<sup>+</sup> Ion in Aqueous Solution

The existence of the  $\rm H_2O^+$  ion in the gaseous state has been fully established by mass-spectrometric observations. From a knowledge of the ionisation potential of the water molecule  $(12\cdot 6~\rm e.v.)^1$  and of other well known thermo-chemical data, it can be shown that  $\rm H_2O^+$  itself should have considerable stability since the association process:

$$H^++OH \longrightarrow H_2O^++6\cdot 1 \text{ e.v.}$$
 (1)

is highly exothermic.

In aqueous systems, however, the situation might be different, since here one has to take into account the hydration energies; under these conditions, therefore, the equilibrium:

$$H_2O^+ aq + H_2O \longrightarrow H_3O^+ aq + OH$$
 (2)

should be of considerable importance. With regard to the position of this equilibrium, it can be said that in view of the structural similarity of the  $\rm H_2O^+$  and  $\rm H_3O^+$ 

<sup>&</sup>lt;sup>1</sup> Cf. W. C. PRICE, Chem. Rev. 41, 257 (1947).

ions respectively, it is to be expected that there will be resonance of the positive charge in both these ions. While this would favour  $\rm H_3O^+$ , it seems reasonable to suppose that this resonance will also confer some stability to the  $\rm H_2O^+$  ion. Furthermore, the dipole interaction energies of a proton with an  $\rm H_2O$  molecule on the one hand and with an OH radical on the other, should be of the same order of magnitude.

In view of these considerations, it is probable that whenever OH radicals are formed in aqueous solution, they may be present to a certain extent, in the form of (hydrated) H<sub>2</sub>O+ ions according to the equilibrium (2).

Hydroxyl radicals are known to play an important part in the reactions of hydrogen peroxide<sup>2</sup> and also in the action of ionising radiations on water<sup>3</sup>.

Recent studies have shown that a number of reactions of OH radicals in aqueous solution exhibit a pH-dependence, which can, in some cases, be accounted for by assuming processes involving the H<sub>\*</sub>O+ ion.

For instance, in the hydroxylation of certain monosubstituted benzene derivatives by OH radicals, it was found that the ratio of the hydroxylated ortho-, para-, and meta-isomers shows a distinct pH-dependence<sup>4</sup>.

The presence of this ion may also have a bearing on the interaction of two OH radicals in aqueous systems. In the gaseous state the experimental evidence is strongly in support of the view that hydrogen peroxide is not formed by the recombination of two OH radicals (2 OH =  $H_2O_2$ )<sup>5</sup>. However, this may be different in aqueous solutions, where it appears that, under suitable conditions, hydrogen peroxide can be formed directly by some interaction of OH radicals. It is suggested that, in acid solutions, hydrogen peroxide formation may take place according to:

$$H_2O^+ + OH \longrightarrow H_3O_2^+$$
 (3a)

$$H_3O_2^+ \longrightarrow H_2O_2 + H^+$$
 (3b)

In general, therefore, in all the reactions of OH radicals in aqueous systems, processes involving the participation of the  $\rm H_2O^+$  radical ion may have to be taken into account.

A detailed study of the pH-dependence of such reactions is now in progress and may give more information regarding the basicity of the OH radial in solution.

J. WEISS

University of Durham, King's College, Newcastle upon Tyne, April 9, 1956.

### Zusammenfassung

Theoretische Überlegungen führen zur Annahme, dass das in der Gasphase wohlbekannte Molekülion  $\rm H_2O^+$  auch in wässeriger Lösung unter geeigneten Bedingungen eine gewisse Stabilität besitzt. Es ist zu erwarten, dass dieses Ion, welches als protoniertes OH-Radikal aufgefasst werden kann, bei Reaktionen in saurer, wässeriger Lösung eine gewisse Rolle spielt.

- <sup>2</sup> Cf. J. Weiss, Advanc. Catalys. 4, 343 (1952).
- <sup>3</sup> J. Weiss, Nature 153, 748 (1944); Brit. J. Radiol. Suppl. 1, 56 (1947).
- <sup>4</sup> H. LOEBL, G. STEIN, and J. WEISS, J. chem. Soc. 1950, 2704; 1951, 405. G. R. A. JOHNSON, G. STEIN, and J. WEISS, J. chem. Soc. 1951, 3275. G. STEIN and J. WEISS, L. chem. Soc. 1951, 3265.
- Soc. 1951, 3275. G. STEIN and J. WEISS, J. chem. Soc. 1951, 3265.

  K. F. BONHOEFFER and T. G. PEARSON, Z. physikal. Chem. [B] 14, 1 (1931).

### PRO EXPERIMENTIS

### A New Fixative for Smoked Kymographic Tracings

By the difficulties encountered in using the traditional fixatives of alcoholic solutions of natural shellacs, we were induced to look for a new synthetic plastic substance for the fixation of smoked kymographic tracings.

The ideal properties for such a surface coating agent are: to be inexpensive, readily prepared, quick in drying and rendering the record permanently soft and flexible without any damaging effect to the smoked surface.

Such a fixative suitable for everyday use can be prepared from commercially available polybutyl methacrylate solutions. The available lacquer¹ contains 50% of the polymer in solution, it is transparent with a pale straw colouring.

As the viscosity of this solution is high, a 1 to 7 dilution of the concentrate is made with dry acetone.

The results obtained with this fixative correspond fully with requirements outlined above.

A. L. DELAUNOIS and T. O. KING

Department of Pharmacology, University of Ghent, Belgium, March 28, 1956.

#### Résumé

Une dilution de polybutyl méthacrylate dans de l'acétone anhydre (rapport 1 à 7) est proposée comme fixateur d'enregistrements sur papier enfumé. Ce nouveau fixateur est incolore, sèche vite, rend les enregistrements flexibles et est peu coûteux.

 $^{1}$  Available as Vinalak No. 5909 from Vinyl Products Ltd., Carshalton, Surrey, England.

### PRO EXPERIMENTIS

# Efficacy of some Histochemical Techniques for Acid Mucopolysaccharides

In an earlier investigation<sup>1</sup> we studied, by chemical methods<sup>2</sup>, the acid mucopolysaccharide content of the rats skin (hyaluronic and chondroitinsulphuric acids) and its variations under diverse experimental conditions. The histochemical examination by metachromasia with thionine was simultaneously made with a portion of the same material, fixed in a 4% formalin solution in 90° alcohol. In accordance with Lison's recommendations<sup>3</sup> for avoiding false reactions, use was made of an aqueous 0.5% solution of that stain in an acid medium (pH 3.2) and of APATHY's syrup for mounting. Some sections were previously incubated for 5 h at 37°C with 1:25,000

<sup>&</sup>lt;sup>1</sup> E. Del Conte, J. Della Sala, and M. Stux, Acta endocrinol. 20, 343 (1955).

<sup>20, 343 (1955).

&</sup>lt;sup>2</sup> R. H. PEARCE and E. M. WATSON, Canad. J. Res. [E] 27, 43

<sup>(1949).

3</sup> L. LISON, Histochimie animale; méthodes et problèmes (Gauthier-Villars, Paris 1936).