

CHEMISTRY

INFLUENCE OF ORGANIC COMPOUNDS ON SOAP- AND PHOSPHATIDE COACERVATES — XIV ¹⁾

The action of fatty acids, alcohols and esters on coacervates of a sulfate soap

BY

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1. *Introduction.*

The influence of many series of organic compounds on soap and phosphatide coacervates have already been investigated by BUNGENBERG DE JONG and collaborators (see H. L. BOOIJ, 1949). For the investigation of the esters, however, there are some objections against the usual substrata.

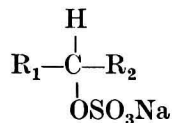
1°. With oleate we work at a high p_H , so saponification may occur and no reliable results will be obtained.

2°. With laurylsulfate we can work at low p_H but to obtain coacervates of this soap we must use a certain amount of menthol or another salt sparing substance which complicates the method (see BOOIJ and VREUGDENHIL, 1950).

3°. Coacervates of cetylsulphate can only be obtained at rather high temperatures (66° C) so that here too saponification may occur (see BOOIJ and VREUGDENHIL, 1950).

So what we want is a sulfate soap (possibility to work at a low p_H) of which coacervates can be obtained at low temperatures without adding some salt sparing substance.

Now all these restrictions are fulfilled in the case of a sulfate soap called T-pol of the following composition: 21 % sodium sec. alkylsulfate, 5 % sodiumsulfate and 74 % water ⁵⁾. The general formula of the sodium sec. alkylsulfate is



¹⁾ Publication no. XIII of this series will be found in Proc. Kon. Akad. Wetensch. Amst. 53, 882 (1950).

²⁾ We are much indebted to the Koninklijke/Shell Laboratorium for a generous gift of T-pol.

³⁾ We wish to express our sincere thanks to the N.V. POLAK & SCHWARZ's Essencefabrieken (Hilversum), who were so kind to send us a great number of esters for our investigations.

⁴⁾ Publication no. 9 of the Team for Fundamental Biochemical Research (under the direction of H. G. BUNGENBERG DE JONG, E. HAVINGA and H. L. BOOIJ).

⁵⁾ We are much indebted to Dr Ir S. L. LANGEDIJK for the informations about the structure and composition of the sample of T-pol.

R_1 and R_2 are alkylgroups with a normal chain, R_1 being in the main CH_3 or C_2H_5 while the total number of carbon atoms of the molecule varies from 8 — 18 (for further particulars see MELSEN, 1950).

Furthermore the solubility in this soap of a large number of esters was rather high which made it easy to test them. The objection against the use of T-pol is, that it consists of a mixture of various carbon chains and *a priori* no micelles with good arrangement, as with oleate, laurylsulfate etc. are to be expected.

We used the following method:

T-pol was diluted 1 : 1 with a sodiumacetate — acetic acid buffer composed of 6 parts 0,2 mol. acetic acid and 94 parts 0,2 mol sodiumacetate ($p_H = 5,8$). This provides us with the so-called "blank" solution. 5 ml of this solution was added, in a calibrated tube, to a mixture of x ml 4,5 n $MgCl_2$ solution and $(10 - x)$ ml aqua dest., whereby x is varied.

Hitherto KCl solutions were commonly used to bring about coacervation, but in the case of the T-pol no coacervation could be obtained with whatever KCl solution, reason why a $MgCl_2$ solution was used. At a certain $MgCl_2$ concentration coacervation appears. Now, five different concentrations of $MgCl_2$ were taken. One whereat just no coacervation appears and further those concentrations that are exactly 0,015 mol/l higher every time. The tubes were kept in a thermostate of 25° for 24 hours; unmixing appears into two layers of which the coacervate layer is the top one.

The concentration of the $MgCl_2$ in the tube is plotted against the volume of the coacervate layer. This supplies us with the so-called "blank" curve. Now the substance whose influence on the coacervation we want to know, is added to the "blank" solution in two different concentrations. Repeating the above experiments, this supplies us with two curves which are shifted with regard to the "blank" one over a certain distance. Now a substance is said to exert a salt-sparing action (here $MgCl_2$ -sparing) if at a certain $MgCl_2$ concentration the volume of the coacervate layer is smaller than in the case of no substance being present. In the reverse case we call a substance to exert a salt-demanding action.

In a diagram the number of mmols substance being present in the tube is plotted against the shift of the curve with regard to the blank, measured in differences in concentration at a coacervate percentage of 60 %.

Before we start the investigation of the esters we will first look at the "components" viz. the acids and the alcohols.

2. Fatty acids.

Acids were investigated from valeric acid up to and including myristic acid. First the influence of hydrochloric and acetic acid were investigated in order to be sure that the established effect could not be due to shifts of the p_H . But no influence at all was found for hydrochloric and acetic acid within the range of concentrations where the investigation took place.

In fig. 1 the action at a concentration of 1 mmol/l is plotted against the number of carbon atoms. It is seen that the action gradually increases with increasing number of carbon atoms. This increase, however, becomes relatively smaller at about eight carbon atoms. If we compare these results with those obtained with oleate (BOOIJ and BUNGENBERG DE JONG, 1949) we find an altogether different behaviour. In the case of the oleate a salt-demanding influence was found, whilst here we find a salt-sparing influence. With oleate, however, a high p_{H} prevailed so that

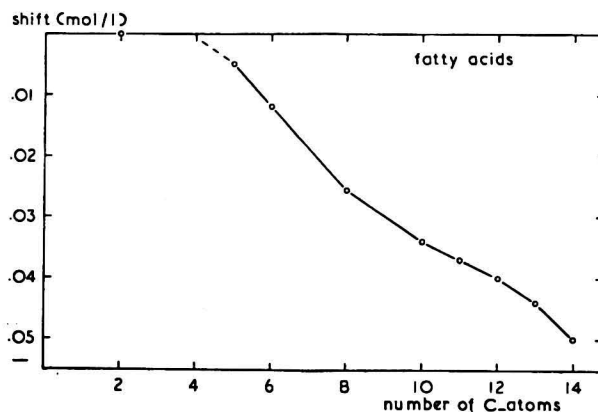


Fig. 1. Action of fatty acids (1 mmol/l) on a coacervate of T-pol. The activity is expressed as the shift of the coacervation curve in mol/l $MgCl_2$.

the fatty acid was dissociated for the most part. Consequently the action of the fatty acid anion was measured whilst we measured the action of the molecule. Moreover the oleate micelle is an ordered one while presumably the T-pol micelle is not, as we have mentioned already. With lauryl- and cetylsulfate a KCl-sparing influence was found (BOOIJ and VREUGDENHIL, 1950). As here too a low p_{H} prevailed the differences in the course of the curves must be sought herein, that the lauryl- and cetyl-sulfate micelles are ordered whilst the T-pol micelle is not.

3. Alcohols.

Again in fig. 2 the action at a concentration of 1 mmol/l is plotted against the number of carbon atoms. Here too it is seen that the action gradually increases with increasing number of carbon atoms, however, only up till 9 carbon atoms whereafter no further increase was observed.

Comparing these results with those obtained with oleate (BOOIJ, VOGELSANG and LYCKLAMA, 1950) and laurylsulfate (BOOIJ and VREUGDENHIL, 1950) we observe for the former that no influence was found from 2 up till and including 4 carbon atoms whereafter an increase in action was observed up till and including 14 carbon atoms; ethyl alcohol, showed a slight salt-demanding action. With the latter, however, ethyl-alcohol also showed a slight salt-sparing action whereafter an increase in

action was found up till and including 12 carbon atoms. The curve reaches its minimum here and proceeds in an upward direction again. We think

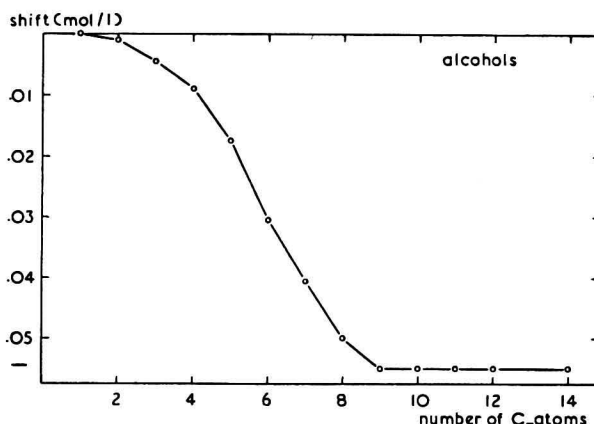


Fig. 2. Influence of normal alcohols (1 mmol/l) on a coacervate of T-pol. Here — as in the case of the fatty acids — we find a salt-sparing action.

that the flat part of the curve with T-pol as substratum finds its explanation in the fact that there is no proper order in the interior of the micelles. A pronounced maximum of activity in a homologous series is only possible if the substratum is very regularly built.

4. Esters.

In fig. 3 the action at a concentration of 1 mmol/l is plotted against the number of carbon atoms. It is seen that with the acetates, butyrates and the esters of ethylalcohol the action gradually increases with increasing number of carbon atoms on the understanding that from about six carbon atoms this increase becomes relatively smaller. The curves

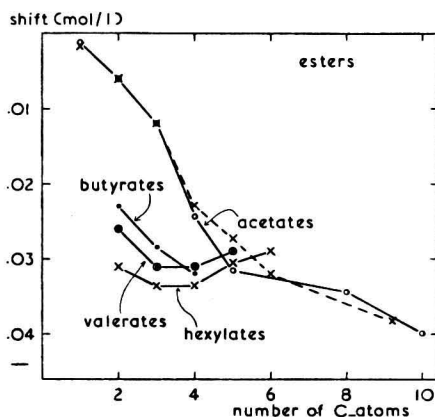


Fig. 3. Action of some series of esters on the T-pol coacervate. The abscissa gives the number of carbon atoms of the alcohol part of the ester. The dotted line shows the influence of the ethylesters (here e.g. 6 = ethylhexylate). Obviously the curves of the acetates and the ethylesters agree rather well.

for the valerates and hexylates show a minimum at 3 — 4 carbon atoms whereafter they proceed again in an upward direction.

The comparison of these esters with the corresponding acids and alcohols may be carried out in two ways viz. we may compare the esters with the acids and alcohols out of which they are composed or we may compare them with those compounds which have the same total number of carbon atoms. If we compare the curves for the esters with the one for the acids we find that they always remain below the latter, which means that the esters all exert a stronger action than the acids out of which they are composed and even than the acids with a same total number of carbon atoms. This is not surprising as the polar nature of the acid group is much stronger than that of the esters.

If we compare the curves for the esters with the one for the alcohols we find that the curves intersect at a value of about 6 carbon atoms. This was to be expected as the curve for the alcohols runs below the one for the acids and moreover the former has a steeper course. The intersection happens at a lower value of the number of carbon atoms for the curve of the hexylates than for the curve of the acetates as a result of the fact that the former displays a minimum at 3 — 4 carbon atoms.

In fig. 4 the action of esters with the same number of carbon atoms at the alcohol as well as at the acid side is plotted against this number. The curve shows a minimum at 4 carbon atoms indicating that the phenomenon

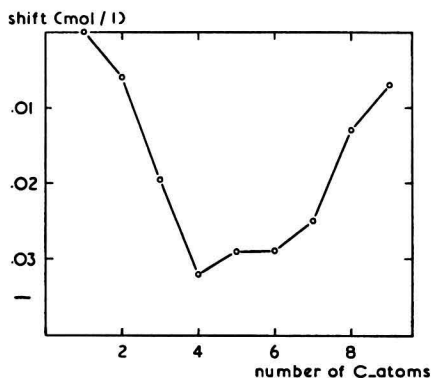


Fig. 4. The action of esters with an equal number of carbon atoms at the acid and the alcohol side of the molecule on a coacervate of T-pol (here 6 means hexylhexylate etc.). A pronounced maximum of activity is observed in this homologous series.

— decrease of the salt-sparing influence with increasing number of carbon atoms — only occurs if the carbon chain has reached a certain length as well at the alcohol as at the acid side of the ester molecule.

A similar series of experiments was carried out with the benzoates, phenylacetates and phenylpropionates, the results of which are plotted in fig. 5. The same phenomenon was observed as with the long aliphatic esters. The minimum is displaced towards lower numbers of carbon atoms

at the alcohol side, going from the benzoates to the phenylpropionates. In accordance with earlier investigations (BUNGENBERG DE JONG, SAUBERT and BOOIJ, 1938) it was found that the benzene ring has the

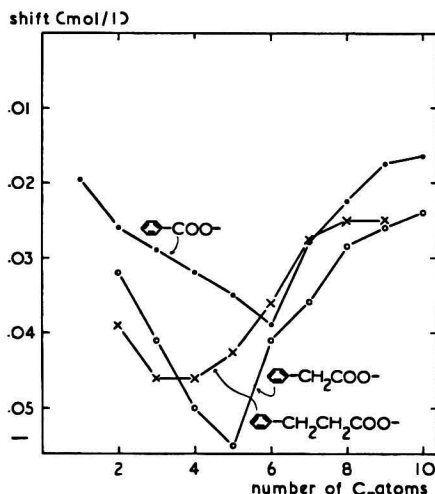


Fig. 5. Influence of esters of aromatic acids on a T-pol coacervate, plotted against the number of carbon atoms of the alcohol chain.

same influence as a normal aliphatic chain of about 4 carbon atoms. For methylvalerate and ethylvalerate exert the same action as respectively methylbenzoate and ethylbenzoate. A cyclohexyl ring has the same influence as a normal aliphatic chain of about 5 — 6 carbon atoms, cyclohexylacetate exerting an influence which is somewhat greater than that of amylacetate. This too is in accordance with earlier experiments (BUNGENBERG DE JONG, SAUBERT and BOOIJ, 1938).

5. Discussion.

The most important phenomenon is the fall in activity with increasing number of carbon atoms, which was found with the higher esters. All the other phenomena which were observed are in accordance with earlier experiments. There are indications (BOOIJ, 1949) that in coacervates micelles are present in which the molecules are arranged in a parallel manner. The salt-sparing influence (of alcohols etc.) is owing to an increase in the London-v. d. Waals attraction forces between the hydrocarbon chains. The salt-demanding influence (fatty acid anions) is due to an increase of the repulsive Coulomb forces of the charged polar groups without a proportional increase of the London-v. d. Waals forces. When a certain amount of the substance is added to the soap solution an equilibrium will arise whereby part of the substance will be taken up by the micelle and part of it will be restricted in the medium. Now when the carbon chain is lengthened one must discern two effects:

With increasing length of the carbon chain the substance becomes

less soluble in water (medium) and so the equilibrium will be shifted in favour of the micelle. In this way we could already explain the ever increasing salt-sparing effect which occurs when the carbon chain is lengthened. A second effect, however, takes place with molecules where the polar groups are very weak or absent. Then it is supposed that these molecules are taken up within the micelle (between the CH_3 -end groups of the carbon chains of the substratum). This may eventually result in a salt-demanding action (BOOIJ, 1949).

A. If an ester has a short and a long carbon chain (e.g. decylacetate) one would suppose that the polar group is in a line with the heads of the soap molecules. In such a homologous series an ever increasing (salt-sparing) effect would be expected. Fig. 6*A* shows how these esters are presumably arranged in the micelle.

The problem arises what place an ester with two long carbon chains will take in the micelle. Both chains will be pushed out of the water and several possibilities (fig. 6*B*, *C* and *D*) are coming to the fore.

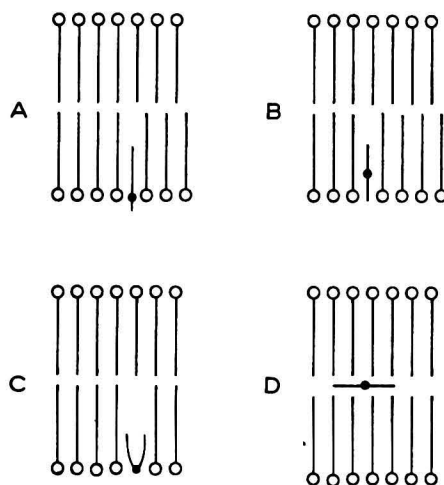


Fig. 6. Possible places of esters within the soap micelle (see text).

B. The molecule is pushed into the micelle, the weak polar group losing its anchorage to the water. It is possible that this would result in a lower salt-sparing action than case *A*.

C. The polar group remains in contact with the water, but the two chains are extended towards the interior of the micelle. It is expected that this would result in a strong decrease of the salt-sparing action as the disturbance of the arrangement in the micelle is very great. This disturbance, however, would become less as the carbon chains of the ester are much lengthened, which would be seen once more as an increase of the salt-sparing activity. This effect in a homologous series (increase of salt-sparing activity, then decrease and finally increase again) we did not find (see fig. 4). Thus scheme *C* does not seem likely to us.

D. It was supposed that aliphatic alkanes — when long enough — would be concentrated in the inside of the micelle. This leads to the view that even a substance with a polar group might go to this place, provided that *a*) the polar group is not situated at the end of the molecule and *b*) that the distance between the polar group and the nearest end of the molecule is at least four carbon atoms. This situation would result in a salt-demanding activity. It might be that the very slight salt-sparing action of long esters which have the polar group in the middle of the molecule (fig. 4) is due to this salt-demanding factor. Then in this homologous series the action would show a maximum, as the partition of the molecules between the places *A*, *B* and *D* (fig. 6) would shift ever more to the right, when the carbon chains of the esters are lengthened.

ALEXANDER and SCHULMAN (1937) have already found with their experiments on the spreading in monolayers of the esters, that an ever increasing energy is necessary to push a longer carbon chain into the water. Long esters viz. cetylpalmitate form a film with the two chains adjacent to one another and both chains standing up from the water surface. This does not implicate that for our experiments schema *C* (fig. 6) is realised as with lengthening of the carbon chain, the anchorage of the water attracting groups is diminished, leading to the possibilities *B* and *D* (see ADAM 1941).

The investigation will be extended with experiments on the speed of saponification of the esters and their spreading in monolayers.

Summary.

1. The influence of fatty acids, normal alcohols and esters on a coacervate of T-pol has been measured. T-pol was selected because with $MgCl_2$ it gives a coacervate at low temperature and at slightly acid p_H .

2. The activity of fatty acids and alcohols increases with increasing number of carbon atoms (up till 9 carbon atoms for alcohols). The fact that the value of the increase diminishes with the higher homologues might be due to the fact that the T-pol micelle is not as regularly built up as e.g. the oleate micelle.

3. In homologous series of esters two different types were found:
a) the activity increases with increasing length of the carbon chain (this was found with esters which have a short and a long chain while the length of the long chain was varied);

b) the activity shows a maximum (this we found with those esters where both chains have at least four carbon atoms).

4. We discussed the possibilities of the situation of the ester molecules in the soap-micelles (fig. 6).

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