

Chemistry. — *Influence of organic compounds on soap and phosphatide coacervates — IX*¹⁾ — *Notes on the action of some organic ring systems with and without polar groups on soap coacervates.* By H. L. BOOIJ. (Communicated by Prof. H. G. BUNGENBERG DE JONG.)

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1. *Introduction.*

The influence of many organic compounds with a polar group on the oleate coacervate has already been measured by BUNGENBERG DE JONG, SAUBERT and BOOIJ (1937, '38) and ROSENTHAL (1939). These experiments were explained by assuming that such molecules exert two opposite forces: a "condensing" action of the hydrocarbon chains and an "opening" action of the polar groups. The picture, however, must be considerably more complex as KOETS and BUNGENBERG DE JONG (1938) showed that aliphatic hydrocarbons do not fit into this picture (for instance *n*-octane having a condensing action at low and an opening action at somewhat higher concentrations). The aromatic hydrocarbons did not show this anomalous behaviour.

BOOIJ and VELDSTRA (1949) investigated the influence of various hydrogenated derivatives of naphthalene acetic acid on an oleate coacervate at high p_H and found that decahydronaphthalene acetate had a much stronger (opening) action than less hydrogenated derivatives of naphthalene acetate. This might be explained by assuming that the decalin nucleus shows a stronger affinity to the apolar part of the oleate micelle than the naphthalene nucleus. The action of these relatively small ions is for a large part determined by the amount of these anions taken up in the soap micelles (cf. the difference between nonylate and undecylate; BOOIJ and BUNGENBERG DE JONG showed that a large percentage of nonylate added to an oleate coacervate remained in the watery medium, whereas undecylate is taken up strongly in the micelles). In the case of decahydronaphthalene acetate (stronger affinity to the non-polar part of the micelles) a greater uptake in the micelles would take place as compared with naphthalene acetate, resulting in a stronger action. This difference led us to the idea to compare the influence of naphthalene and decalin (which, however, proved to be reverse; see section 2). After that it seemed worth while to investigate the action of other nuclei (especially heterocyclic ones).

¹⁾ This series was published under the name: "Einfluss organischer Nichteletrolyte auf Oleat- und Phosphatidkoazervate" in *Protoplasma* **28**, 498, 543 (1937); **29**, 481, 536; **30**, 1, 53, 206 (1938), and under the name: "Influence of organic compounds on oleate and phosphatide coacervates" in *Bioch. Bioph. Acta* **3**, 242 (1949).

2. *On the action of some aromatic, alicyclic and heterocyclic nuclei.*

To 70 ml oleate solution (10 g Na-oleate + 525 ml H₂O + 25 ml KOH 5 mol) the desired quantity of the organic substance is added, after which the solution is gently rolled in order to dissolve the added compound. Sometimes this will take a whole day. Then varying amounts of KCl were added according to the following scheme:

$$\left\{ \begin{array}{l} 5 \text{ ml oleate solution} \\ x \text{ ml KCl } 3.8 \text{ n} \\ (14 - x) \text{ ml H}_2\text{O.} \end{array} \right.$$

The mixtures were thoroughly shaken and set in a thermostate of 25° C. Next day the volumes of the coacervates formed were read and plotted against the KCl concentration (see for the method also BOOIJ and BUNGENBERG DE JONG, 1949). Organic substances shift this KCl-curve and this shift — expressed in KCl concentration (mol/l) — is taken as a measure of the action of the substance added. The shift may go to lower KCl concentrations (—; “condensing action”) or to higher (+; “opening action”).

The condensing action of decalin appears to be appreciably smaller than that of naphthalene and tetralin (fig. 1). One would not expect this

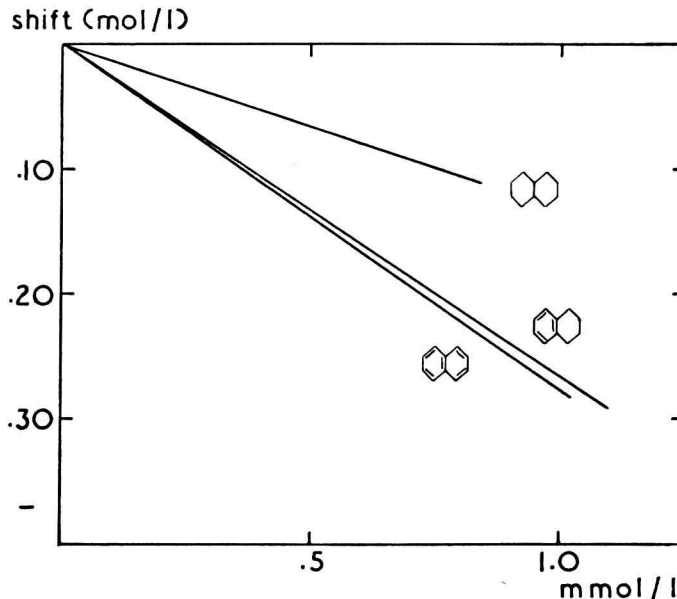


Fig. 1. Condensing action of naphthalene, tetralin and decalin on an oleate coacervate. Abscis = concentration of added substance, ordinate = shift of the coacervation curve expressed in KCl concentration.

after the experiments of BOOIJ and VELDSTRA (1949). They explained the difference between decahydronaphthalene acetate and naphthalene-acetate by supposing that the former compound is taken up more readily by

the micelle as the affinity of the decalin nucleus to the apolar part of the oleate micelle is somewhat greater than that of the naphthalene ring system. In the case of naphthalene and decalin, however, one may safely assume that both substances are taken up to a large extent into the micelles and it is not yet clear why the situation is exactly the reverse of that with the acetates.

With heterocyclic ring systems the results are easily explained. One would suppose that the exchange of a carbon atom of an aromatic ring for another atom will result in a decrease of the action. This decrease will be greatest with N, less with O and still less with S. Generally speaking the aromatic character of the resulting heterocyclic ring grows stronger in this direction. Especially the NH group will give the compound a more polar character (for instance furane and thiophene resemble benzene, while pyrrol is to be compared with phenol). Fig. 2 and 3 demonstrate this clearly. Introduction of a N-atom into a ring results in a marked decrease of activity (cf. indole — indene; quinoline and isoquinoline — naphthalene).

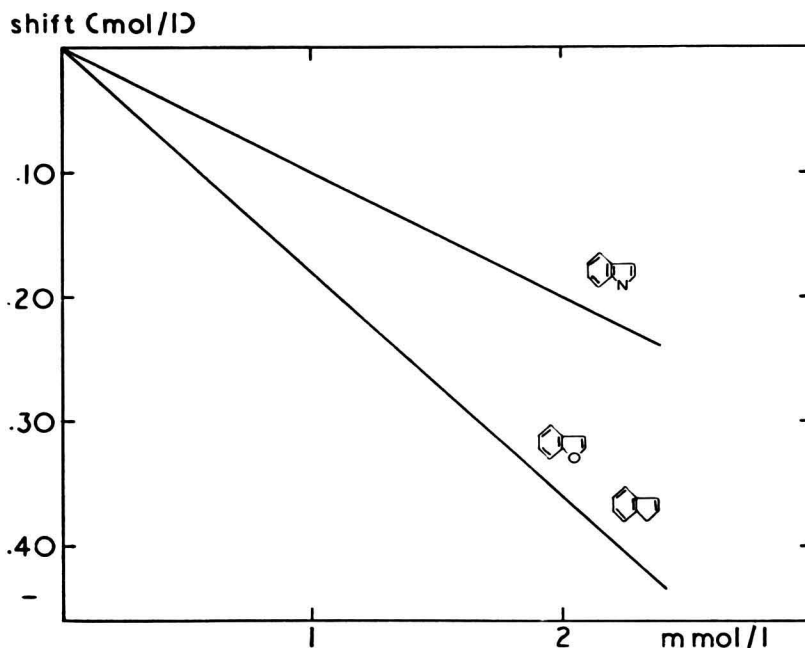


Fig. 2. Influence of heterocyclic ring systems (coumarone and indole) on an oleate coacervate as compared with that of indene.

This decrease is much smaller or absent with the O-atom (cf. coumarone-indene). We got the impression that furane has a smaller activity than thiophene, but we could not prove that beyond doubt. The difference between acridine and diphenyleneoxyde was easily demonstrated. Pyridine and piperidine distinguish themselves markedly (fig. 4). If it may be assumed that at high p_H (we worked at a p_H 12–13) the polar groups

are not dissociated, the most important difference of these molecules would be their aromatic or alicyclic character. Then here too the substance with the alicyclic ring has the strongest action. This would lead to the idea that

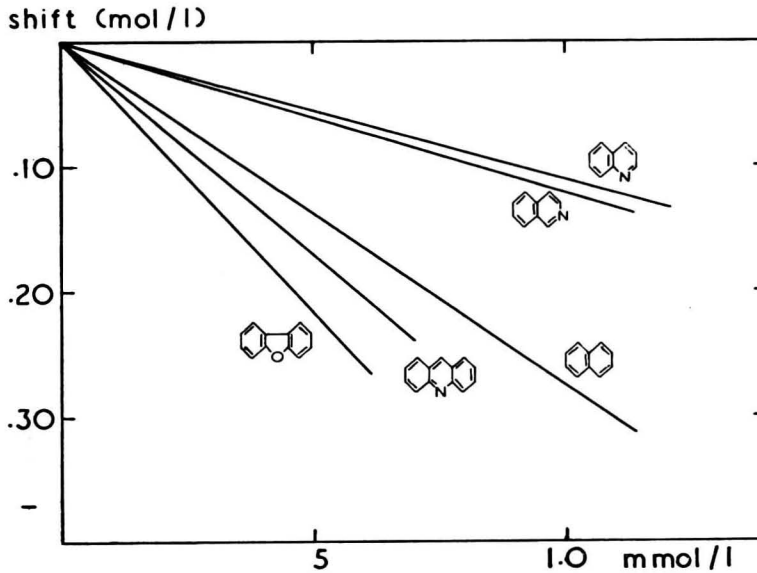


Fig. 3. The action of quinoline, isoquinoline, acridine and diphenyleneoxyde on the oleate coacervate as compared with naphthalene.

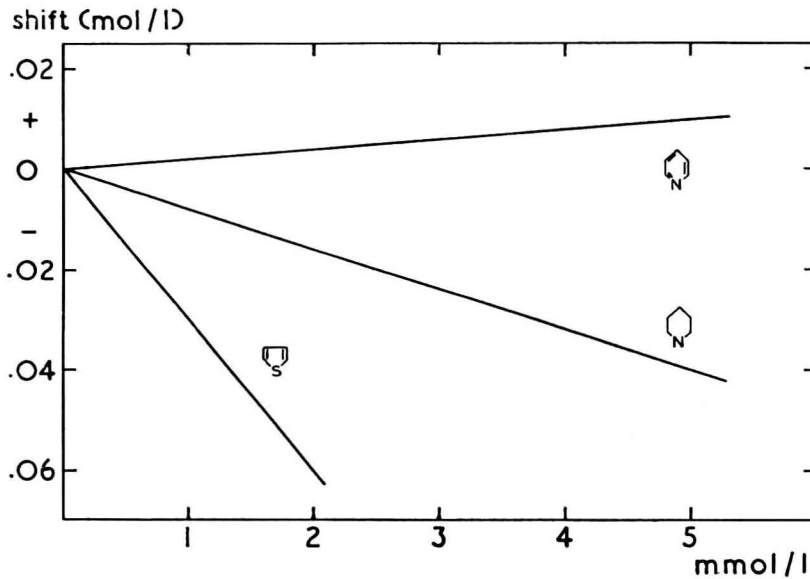


Fig. 4. Influence of pyridine as compared with piperidine and of thiophene on the oleate coacervate. Note that the action of these small molecules is much less than that of bigger molecules. (An opening action (pyridine) is expressed by the sign +, a condensing action by —.)

a substance with an alicyclic ring has the strongest action provided a polar group is part of the molecule. When the compounds have no polar groups (fig. 1) the reverse will be true.

Finally we found that imidazole, piperazine and pyrazole had no influence on the coacervate in the concentrations measured (up to 1 mmol). Here it may be supposed that the substance added will be found practically exclusively in the watery medium, so that the action on the micelles is very slight.

3. The influence of introduction of a polar group.

First we measured the action of the isomeric methylcyclohexanols as compared with cyclohexanol and cyclohexanon (fig. 5). The place of the methylgroup influences the action, the longest molecule having the greatest

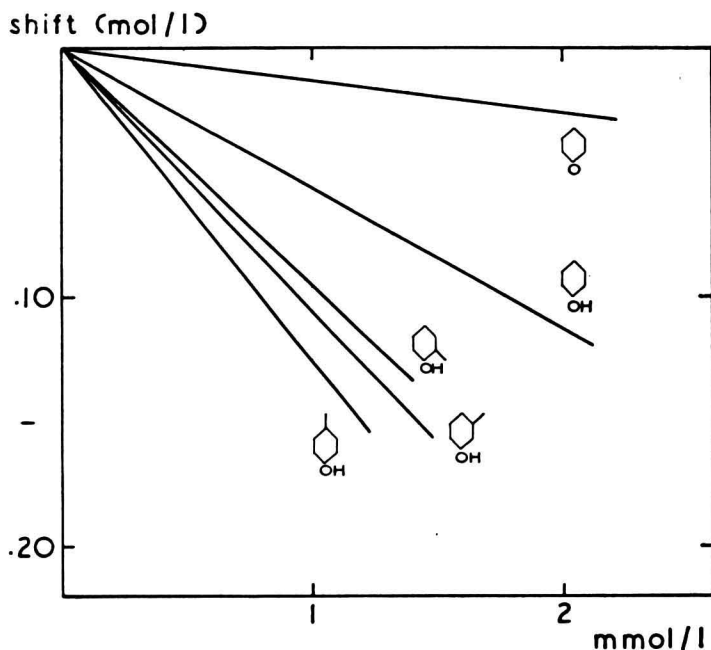


Fig. 5. The action of ring systems with a polar group depends on the nature of the polar group (cyclohexanon < cyclohexanol), the hydrophilic-hydrophobic balance (cyclohexanol < methylcyclohexanol) and the length of the molecule (see the difference between the isomeric methyl cyclohexanols).

activity. It had been found already that the keton-group diminishes the "condensing" action if introduced in stead of an OH-group (BUNGENBERG DE JONG et al., 1938).

These results were in accordance with former publications in this field. An investigation of terpenes and related substances showed a somewhat surprising phenomenon (fig. 6). This fact was the relatively low condensing activity of pinene and d-limonene. A priori one would not expect that

menthol, borneol and terpineol have a stronger action than the apolar substances mentioned. If we would try to explain these facts along the lines

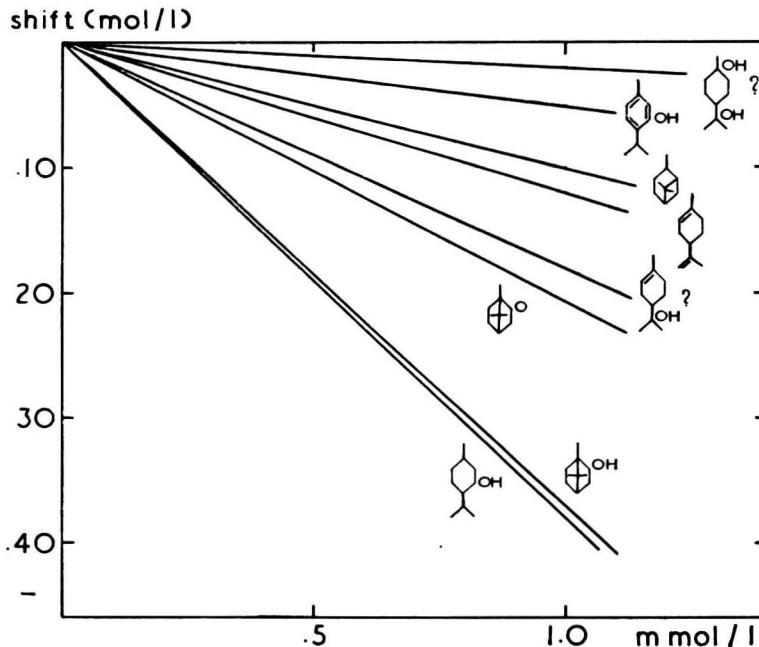


Fig. 6. The action of pinene, *d*-limonene, camphor, borneol, menthol, thymol, terpineol (mixture of isomers) and terpinhydrate (mixture of isomers) on an oleate coacervate.

formerly developed, it would be necessary to assume that a double bond exerts a stronger opening action than an OH-group, which is contrary to many known facts. So it seems evident that another factor plays a part here.

Many facts have been found to be exactly as expected. Obviously borneol has a much stronger activity than camphor (difference of alcohol and keton-group). Introduction of a second OH diminishes the action very markedly (see terpinhydrate). The difference between thymol and menthol must be ascribed to the high p_H at which these experiments were performed. Then the aromatic OH-group will be present in the dissociated phenolate form, which must result in a much smaller action.

4. Experiments on a coacervate of cetylsulfate.

It might be possible that the difference in action of naphthalene and decalin (see fig. 1) is based on a strong affinity of the aromatic nucleus to the double bond of the oleate molecule. Then a coacervate of a soap with a saturated carbon chain should show the reverse phenomenon. Obviously, this explanation would not help us to see why an alicyclic nucleus with a polar group does act more strongly than a similar aromatic derivative. An

experiment with a coacervate of cetylsulfate showed that the phenomenon is in that case exactly the same (fig. 7).

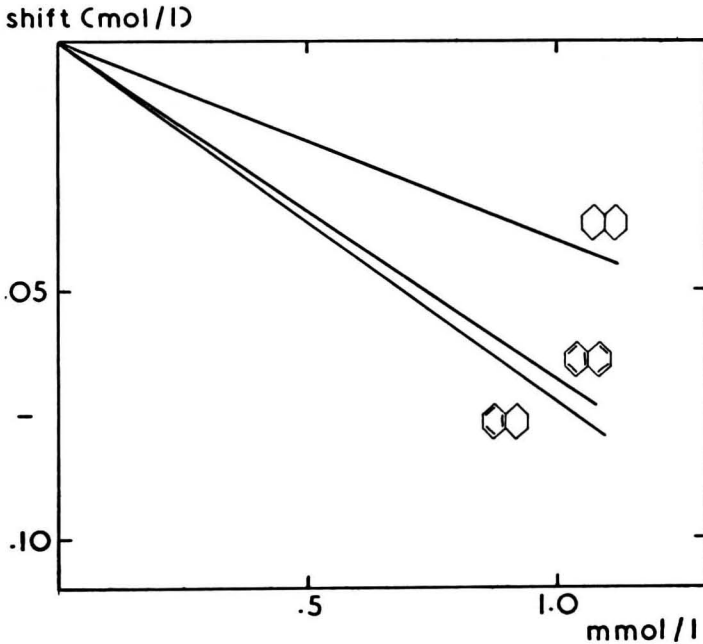


Fig. 7. The influence of naphthalene etc. on a coacervate of cetylsulfate is comparable to that on an oleate coacervate. Here the activity is expressed in the shift of the NH_4Cl curve.

One must work at a high temperature to coacervate a solution of cetylsulfate. The method we used was as follows: 5 g Na-cetylsulfate was dissolved in 200 ml distilled water and kept at a temperature above 60°C , as otherwise the soap will crystallize. Here too, as in the case of our experiments with oleate, a certain amount of the substance to be measured is added to a known volume of the soap solution (20 ml) and dissolved. Then we make the following mixtures:

$$\left. \begin{array}{l} x \text{ ml } \text{NH}_4\text{Cl } 5 \text{ n} \\ (7 - x) \text{ ml } \text{H}_2\text{O} \\ 3 \text{ ml cetylsulfate solution.} \end{array} \right\}$$

The cetylsulfate is added to the salt-water mixture when the latter has reached the temperature of the thermostat in which the experiments are performed (66°C). These mixtures are well shaken and because of the high temperature heavy test tubes with ground glass stoppers have been used. Next day the volumes of the coacervates formed are read and plotted against the concentration of NH_4Cl . As usually, the shift of the coacervation curve under the influence of the added substance is taken as a measure of the activity of that substance.

Fig. 7 shows that the relation between naphthalene and decalin is exactly as with the oleate coacervate. This means that the double bond in the oleate molecule does not play an important part as regards this phenomenon.

We had the opportunity to investigate with this coacervate some other questions which arose during our experiments with oleate coacervates. The relation between borneol and camphor (fig. 8) is comparable with that on

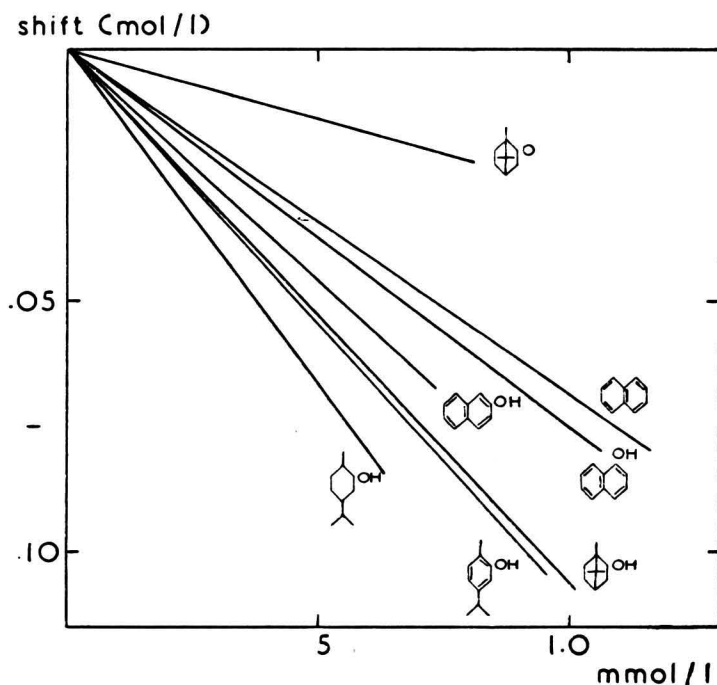


Fig. 8. Activity of α and β naphthol, naphthalene, camphor, borneol, menthol and thymol with a cetylsulfate coacervate as substratum. Note the much more pronounced activity of thymol as compared with that on oleate (fig. 6).

the oleate coacervate (fig. 6). Here too the keton group diminishes the condensing activity. As in this coacervate the p_H is in the neighbourhood of the neutral point (NH_4Cl was used as salt, while no KOH was added to the soap) one would expect that thymol and menthol will not show such diverging actions as with oleate (cf. fig. 8 and 6). This proved to be the case, menthol having a slightly stronger action, in agreement with the rule that an alicyclic compound with a polar group has a stronger action than the related aromatic one. Experiments on the naphthols gave us one more example of substances with a polar group with a stronger action than the parent substance (cf. the examples menthol, terpineol etc. already mentioned; there it was not possible to compare them with their real parent substances). Of course, β naphthol is somewhat more active than α naphthol, as its molecule is somewhat longer (see also BOOIJ and VELDSTRA 1949,

page 265 for the difference between naphthalene (1) and naphthalene (2) derivatives).

5. Discussion.

Two main points may be derived from our experiments:

a) The difference between alicyclic and aromatic nuclei is rather complicated, as the action depends on the fact whether a polar group is bound to the molecule. Without polar group the aromatic nucleus has a stronger action than the similar alicyclic ring, while with a polar group the alicyclic derivative is the stronger one. The phenomenon that an alicyclic ring without a polar group has a lower condensing action than the comparable aromatic one seems to be a general rule. For example, *o*-xylyol and dimethylcyclohexane behave in the same manner (fig. 9).

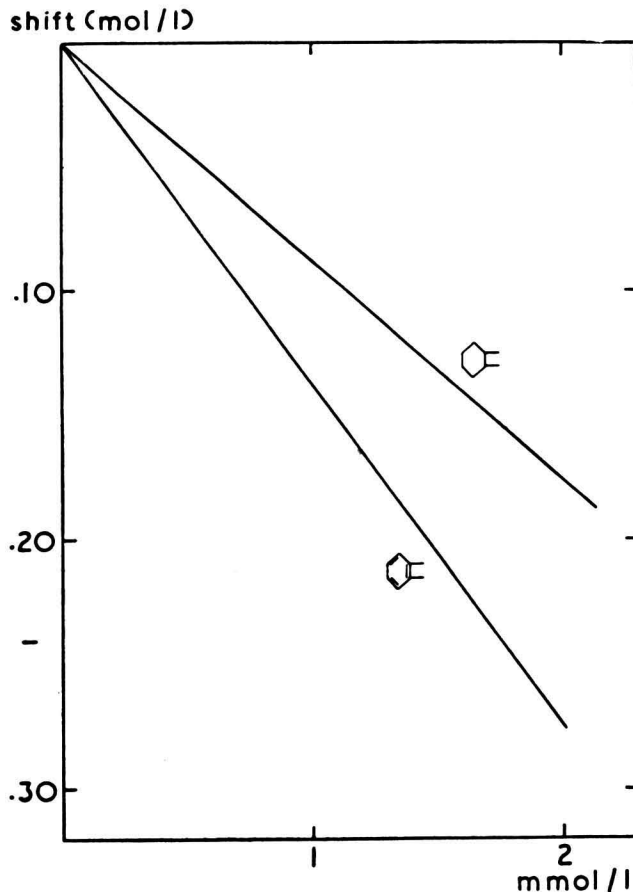


Fig. 9. The action of dimethylcyclohexanol on oleate as compared with that of *o*-xylyol.

b) Some substances with a polar group exert a stronger action than their parent substances (cf. naphthol and naphthalene), a fact which is not in accordance with rules formerly laid down.

Several facts led to the conclusion (BOOIJ, 1949) that organic molecules may act in two ways on an oleate micelle. A substance like benzene might act *a*) in between the parallel carbon chains of the soap molecules and *b*) it might form a layer between the CH₃ end groups of these molecules resulting eventually in the well known „sandwich” micelle of Hess et al. (1941).

When we ask ourselves what molecular properties would give a preference to action *a* or *b*, we may start from the action of fatty anions on the oleate coacervate (BOOIJ and BUNGENBERG DE JONG, 1949). This action can only be explained by assuming that the fatty anions added will be arranged parallel to the carbon chains. This seems logical as strong forces will oppose the introduction of the charged polar group into the apolar inside of the micelles. An investigation of the homologous series of normal alcohols ²⁾ led to the same conclusion as regards the OH group. Obviously, in these cases the polar group serves as a kind of anchor, holding the molecule in the position between the parallel carbon chains of the soap molecules.

The logical consequence is that without that anchor the added substance may be found between the planes of the CH₃-groups. So hydrocarbons might be expected to concentrate in the latter place. If we now see that *n*-octane (KOETS and BUNGENBERG DE JONG, 1938) shows at low concentrations a condensing and at higher ones an opening action, then we might assume that the concentration of *n*-octane within the inside of the micelle results in an “opening” action. The mechanism of this action (which must be distinguished from the opening action of fatty anions) is far from being understood.

As a working hypothesis we may adopt, however, that the action of hydrocarbons depends on their distribution within the micelle, two places (between the carbon chains and between the CH₃-planes) being available. Analogies with the action of compounds with a polar group then teach us that concentration at the former place gives a “condensing” action ³⁾ while the added substance in the latter place acts as an “opening” compound. In the light of this hypothesis the behaviour of alicyclic and aromatic ring systems loses its enigmatic character.

The aromatic nuclei may be seen as somewhat more “polar” than similar alicyclic rings. Thus the alicyclic rings (without polar groups) tend to concentrate more in the inside of the micelles, in other words they have a less pronounced “condensing” action than the aromatic rings. When, however, they are provided with a polar group like OH etc. this tendency is annihilated and the stronger affinity of the alicyclic ring to the apolar chains of the oleate comes to the fore. Here the “condensing” action will be stronger than with comparable aromatic ring systems.

Our working hypothesis thus throws light on the two points mentioned

²⁾ Not yet published.

³⁾ With the exception of long fatty anions.

at the beginning of this section. The difference between alicyclic and aromatic rings can easily be understood. Introduction of a polar group into a ring system provides a strong preference for the places between the carbon chains and prevents the molecule from going to the inside of the micelle and exerting its "opening" action. With the parent substance the action was the sum of a condensing and opening action, resulting from a distribution equilibrium between the two places available. It is to be expected that this equilibrium will have a different value for various hydrocarbons.

My sincere thanks are due to Prof. H. G. BUNGENBERG DE JONG and Dr H. VELDSTRA. It is doubtful whether the theoretical explanation of the phenomena described would have been developed without the many discussions we had on this subject. From the Research laboratory, Combinatie N.V. en Amsterdamsche, Bandoengsche en Nederlandsche Kininefabrieken I received a considerable quantity of cetylsulfate.

Summary.

1. With an oleate as well as a cetylsulfate coacervate it was observed that aromatic ring systems (without polar group) have a greater "condensing" action than comparable alicyclic ones.
2. For compounds with a polar group the situation is exactly reversed.
3. Heterocyclic rings will arrange themselves according to their aromatic character, the activity increasing in the range $N < O < S < C$.
4. In some cases the introduction of a polar group results in a stronger activity than in the parent substance (e.g. naphthol - naphthalene).
5. Starting from the working hypothesis that apolar substances have essentially two places of action within the soap micelles (a. in between the parallel carbon chains and b. between the CH_3 planes) the phenomena described have been explained. A compound with a polar group is held in position a., for substances without a polar group an equilibrium between the amounts present at a. and b. will establish itself. Obviously, this equilibrium will depend on the molecular structure and presumably on the situation in the micelle.

LITERATURE.

1. BOOIJ, H. L., in KRUYT'S Colloid Science II, Elsevier, Amsterdam 1949, Chapter XIV.
2. BOOIJ, H. L. and H. G. BUNGENBERG DE JONG, *Bioch. et Bioph. Acta* **3**, 242 (1949).
3. BOOIJ, H. L. and H. VELDSTRA, *Bioch. et Bioph. Acta* **3**, 260 (1949).
4. BUNGENBERG DE JONG, H. G., G. G. P. SAUBERT and H. L. BOOIJ, *Protoplasma* **30**, 1 (1938).
4. HESS, K., H. KIESSING and W. PHILIPPOFF, *Fette und Seifen* **48**, 377 (1941).
6. ROSENTHAL, S., Thesis, Leiden (1939).

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