On the Poisonous Action of Alcohols upon Different Organisms.

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The physiological action of different alcohols has been investigated by various authors, but the conclusions reached have not been always concordant. According to Dogiel,[1] for instance, methylic alcohol is less poisonous than ethylic alcohol, while Dujardin-Beaumetz and Andigé[2] state just the reverse as the result of their experiments. A. Schneegans and J. v. Mering[3] conclude that the primary alcohols are less narcotic than the secondary ones, and the latter less so than the tertiary, but this does not agree with the results of the experiments made by W. Gibbs and E. J. Reichert.[4] Nevertheless it may be concluded from many different experiments that on the whole the toxic action of alcohols in the methylic series runs parallel with the increase in the number of carbon atoms contained in their molecule. However, the subjects serving for these experiments were chiefly warm-blooded animals, and very rarely the lower forms of life. It seemed, therefore, of some interest to compare in this respect *some representatives of all kinds of living organisms.* Of special interest also it

1. *Pflüg. Arch.*, **8,** 605; or *Americ. Chem. Jour.*, **13,** 370.

2. *Jour. Chem. Soc. Lond.*, **30,** 539, from *Compt. rend.*, **83,** 80–82. In the *Americ. Chem. Jour.*, **13,** 370, and *Jour. Chem. Soc. Lond.*, **60,** 1393, they are said to have found that "methylic alcohol was *less* poisonous than ethylic alcohol, but it is evident from the abstract in J. Ch. Soc. Lond., **30,** 539 that they have been misquoted.

3. *Chem. Centralb.*, 1892, II., 367.

4. *Americ. Chem. Jour.*, **13,** 370.

seemed to be to compare together the effects of methylic and ethylic alcohols. I have examined altogether the action of nine alcohols: methylic, ethylic, normal and iso-propylic, normal, iso-, and tertiary butylic, normal amylic alcohol, and, finally, allylic alcohol (CH: CH. CH_2. OH). These alcohols were diluted with distilled water[1] to the extent stated in connection with each experiment, the percentage given being that by volume.

Action of the Alcohols upon Lower Vertebrate Animals.

For the experiments were chosen tadpoles of *Bufo vulgaris*, Laur, in such a stage of development that the hind legs had made their appearance. Three individuals were put into 50 c.c. of the alcoholic solution of a certain strength. Control experiments with plain water were made in every case. In 0.1% solutions they became motionless after $1\frac{1}{2}$–2 hours in allylic alcohol; after 10–25 minutes in amylic alcohol; and after one hour in butylic alcohol; while those in the other 6 alcohols of the same dilution were apparently not injured. After 24 hours those in the butylic alcohol had almost entirely recovered, but those in the amylic and allylic alcohols were dead. The tadpoles in the other 6 alcohols were alive after 10 days.[2]

With methylic and ethylic alcohol the experiments were still further extended. In 0.3%, 0.5%, 0.7%, and 1% solutions the intensity of the narcotic action increased with the degree of the concentration, as was to be expected, but in all cases the action of ethylic alcohol was stronger than that of methylic alcohol. Further, in 1.5% ethylic alcohol all the tadpoles showed great stupor, while in methylic alcohol of the same strength one only of the three was

1. Only in those cases where infusoria served for the experiment, was ordinary well water used.

2. In all cases some *Spirogyra* threads were added as food for the animals.

rendered insensible, the other two were still moving after one hour. After 24 hours, however, all of them recovered, those in ethylic alcohol being much weakened, while those in methylic alcohol were almost normal. Tadpoles in 2% solution of ethylic alcohol ceased to move in 10 minutes and after 24 hours two of them died, while a third, although apparently recovered, died 40 hours later. A 2% solution of methylic alcohol proved to be much weaker in action upon them than ethylic alcohol. All were apparently insensible in from 20–50 minutes, but only one of them died after 24 hours, while the other two recovered, and were still alive 40 hours after. In 2.5% solutions, however, they were insensible in from 4–18 minutes, and died soon afterwards.

In further experiments with propylic alcohols (normal and iso) we found that:—in 0.5% solutions the tadpoles stopped their motion within 40 minutes in the normal and within 30 minutes in the iso-propylic alcohol, and after 24 hours two of them recovered in each case, while the others died. In 0.7% solutions motion ceased after 17–30 minutes immersion in the normal and after 9–20 minutes in the iso-alcohol. After 24 hours, however, although those in the normal alcohol were dead, two in the iso alcohol partly recovered, only one out of the three dying. In 1% solutions of these alcohols, the animals became insensible within 10 minutes, and died soon afterwards. In the case of the butylic alcohols insensibility set in within 5 minutes in 0.3% normal butylic, after 3–9 minutes in 0.5% isobutylic, and after 5–15 minutes in 0.7% tertiary butylic alcohol, death following in all these cases within 24 hours. Insensibility was also established after 9–14 minutes in 0.3% isobutylic alcohol and after 9–20 minutes in 0.5% trimethylcarbinol, but in both cases the animals recovered, though those in the former alcohol seemed much more prostrated than those in the latter.

It would seem from the preceding experiments with 0.1 % amylic and allylic alcohols that the toxicity of amylic alcohol upon the tadpole is stronger than that of allylic alcohol, contrary to what has been found in experiments upon other forms of life which will be described later on. I made, therefore, further experiments with the two alcohols, of the same dilution (0.1 %), as well as of higher dilutions. In 0.1 % solutions tadpoles again died sooner in amylic than in allylic alcohol. When the animals however appeared to be insensible they were this time removed into fresh water; whereupon those that had been in the amylic alcohol recovered, while those from the allylic alcohol did not; the latter also had convulsions, but not the former. In 0.05 % amylic alcohol solution most of the tadpoles were paralysed in $1\frac{1}{2}$ hours, but after 18 hours they recovered, and were alive after 5 days. In 0.01 % solution of that alcohol hardly any action could be noticed. In other experiments, the animals were killed in—

0.05 %	allylic	alcohol	after	2	hours
0.01 %	,,	,,	,,	$4–5\frac{1}{2}$	,,
0.005 %	,,	,,	,,	$5–6\frac{1}{2}$	,,
0.002 %	,,	,,	,,	$6–6\frac{1}{2}$	,,
0.001 %	,,	,,	,,	$6\frac{2}{3}–7$	,,
0.0005 %	,,	,,	,,	8	,,
0.0001 %	,,	,,	,,	12–15	,,

These observations show that allylic alcohol is a *very strong poison*, far stronger than amylic alcohol.

Action upon Lower Aquatic Animals.

Experiments were carried out upon Ostracodes (*Cypris* and *Cypridina*) and Infusoria (principally *Paramœcium*) in the same manner as those described above. Allylic alcohol was applied in solutions of

0.1–0.005 % and even in the latter case all life was extinct within 24 hours. Amylic alcohol in 0.1 % killed all ostracodes and many infusoria within one day, though some infusoria were found alive even after 5 days, but by a solution of 0.5 % all animals without exception were killed after one day. In butylic alcohol of the same strength most of the animals died after two days, and in isobutylic alcohol after 3 days; the few individuals that still showed signs of life were evidently more or less paralysed.[1]

In 1 % solutions of normal butylic, isobutylic, and tertiary butylic alcohol (trimethylcarbinol), and of isopropylic and propylic alcohols, infusoria were found dead after 18 hours, ostracodes after 18 hours in isopropylic and butylic alcohol, after two days in isobutylic alcohol, and after three days in propylic alcohol. In tertiary butylic alcohol most ostracodes were dead after two days, but some individuals still showed convulsions two days later.

The observations made concerning the activity of methylic and ethylic alcohol are contained in the following table :—

Alcohol.	1 %	3 %
Methylic $CH_3 \cdot OH$.	Infusoria and Ostracodes were seen alive after 4 days.	Strong narcotic action observed after 20 hours; no life after 4 days.
Ethylic $CH_3 \cdot CH_2 \cdot OH$.	Ditto[2]	Strong narcotic action after 4 hours; all life ended after 20 hours.

1. In another experiment the animals died sooner, the temperature, which, of course, has great influence, being higher. In the experiments with solutions of normal and isopropylic alcohols of the same dilution, the animals were found alive after four days.

2. Compare O Lœw, *Natürl. System der Giftwirkungen*, S. 26.

Action upon Phænogams.

The first series of experiments was carried out with seeds which were just beginning to germinate after having been soaked in water. In each experiment 12 seeds were left in 50 c.c. of the diluted alcohol for 24 hours in a closed vessel; after pouring off the liquid the covered vessel was left at the ordinary temperature and the further development of the germ observed. The seeds treated were those of barley, *soya* bean, Swedish turnip, Japanese onion (*Allium fistulosum*, L.), and *shungiku* (*Chrysanthemum coronarium*, L.). In one set of experiments solutions of 0.1% of the alcohols were used, while in another set barley seeds were treated with 1% and turnip seeds with 0.5% and 1% solutions. The results are shown in the following table:—

Alcohol.	0.1%	0.5%	1%
Methylic $CH_3 \cdot OH$.	Alive.	Alive.	Alive.
Ethylic $CH_3 \cdot CH_2 \cdot OH$.	Alive.	Alive.	Alive.
Propylic $CH_3 \cdot CH_2 \cdot CH_2 \cdot OH$.	Alive.	Alive.	Barley mostly, turnip all killed.
Isopropylic $CH(CH_3)_2 \cdot OH$.	Alive.	Alive.	Barley mostly, turnip all killed.
Butylic $CH_3 \cdot (CH_2)_2 \cdot CH_2 \cdot OH$.	Alive.	Dead.	Dead.
Isobutylic $CH_3 \cdot (CH_3)_2 \cdot CH \cdot OH$.	Alive.	Dead.	Dead.

Alcohol.	0.1%	0.5%	1%
Tertiary butylic $C(CH_3)_3 \cdot OH$.	Alive.	Alive.	Barley mostly, turnip all killed.
Amylic[1] $CH_3 \cdot (CH_2)_3 \cdot CH_2 \cdot OH$.	Only turnip mostly injured.	Dead.	——
Allylic $CH_2 : CH \cdot CH_2 \cdot OH$.	All seeds killed.[2]	——	——

In experiments with methylic and ethylic alcohols of higher concentrations, of 2% and 3%, I found that while 2% ethylic alcohol had killed in 24 hours the turnip germs, methylic alcohol had not killed them even when of the strength of 3%.

In other experiments the action of propylic alcohol was compared with that of allylic alcohol on young *soja* beans which had reached the height of 15 cm. in water-culture. One plant was placed in 500 c.c. of 0.1% solutions of each alcohol. That in the allylic alcohol died on the 3rd day, while that in the propylic alcohol was not injured. A similar experiment was carried out upon pea plants about 35 cm. long. The lower three leaves of the plant placed in allylic alcohol (0.1%) turned yellow and dried up in 3 days, and its upper leaves in 5 days; and the plant itself was found dead in 7 days. In a control experiment, also no injurious effect was observed in the case of propylic alcohol.

1. In the case of amylic alcohol turnip seeds were also killed in the 0.3 % solution.

2. In a few turnip seeds the cotyledons showed some development, but in none did the rootlets of the embryo show any.

Action upon Algæ.

In 0.1% solution of allylic alcohol *Spirogyra communis* was killed in 24 hours, while in the solutions of the other alcohols of the same dilution it was found healthy even after 10 days. In other experiments, allylic alcohol of 0.05% also killed *Spirogyra* in 24 hours. Allylic alcohol of 0.01% killed it in three days, but 0.005% had no effect even in 10 days. In 0.5% solutions, this alga was killed by amylic alcohol in one day, by butylic in 3 days, by isobutylic in 4 days, still later by tertiary butylic, propylic, and isopropylic alcohols. In dilutions of 1%, methylic and ethylic alcohols had no injurious effect, but propylic alcohol killed the cells within 3 days, and isopropylic, butylic, isobutylic, and tertiary butylic alcohols killed it within 2 days.[1] In 2% solution, ethylic alcohol proved much more injurious than methylic, for while most of the cells were dead after 3 days in the former, only a few were so in the latter, but in either alcohol all the cells were dead after 5 days. In a 3% solution of ethylic alcohol all were killed within 3 days, while in that of methylic alcohol they lived 4 days. In a 4% solution of methylic alcohol, however, all the cells were killed within two days.

Action upon Microbes.

For these experiments principally methylic, ethylic, amylic, and allylic alcohols were used. In one series of experiments their toxical actions were tested and in other series their nutritive qualities. One drop of putrefying broth was introduced into the alcoholic solution of a certain dilution, and then a sterilised solution of meat-extract was infected from the alcoholic solution after this had stood 24 hours.[2] The results are shown in the following tables :

1. In another case 1% isopropylic alcohol solution killed *Spirogyra* cells within one day.
2. Of course, control experiments with plain water were also made.

I.

Alcohol.	0.1 %	0.5 %	1 %
Amylic	Strong development at the same time as in the control experiment.	The same as with 0.1 %	Development two[2] days later than in the control expt.
Allylic	Development one day later than in the control experiment.	Weak development[1] 4 days later than in the control expt.	Weak development 4 days later than in the control expt.

II.

Alcohol.	5 %	15 %	20 %
Methylic and Ethylic	No difference from the control experiment.	Much less development than in the control expt.	No development after 10 days' standing.

In a 2 % solution of amylic alcohol bacterial life is evidently greatly depressed, for the infection of meat-extract solution with this alcoholic solution did not induce any development within 7 days.

In a second series of experiments 0.1 % potassium phosphate, 0.01 % magnesium sulphate, and the alcohol to be tested in a certain dilution, were added to a 0.5 % meat-extract solution. The infection was made from putrid meat. The results are as follows :—

0.1 % allylic alcohol : after 6 days, a weak bacterial development.

0.5 % ,, ,, : after 8 days, very slight development.

1. The bacterial vegetation consisted principally of one kind of thin long bacilli.
2. Here principally micrococci were noticed.

1% amylic alcohol : after 4 days, considerable development.
10% ethylic „ : after 14 days, large development.
15% „ „ : after 14 days, quite clear, free from bacteria.
20% „ „ : no development.
10% methylic „ : strong development.
15% „ „ : weak development.
20% „ „ : no development.

In a third series of experiments no source of carbon for the growth of bacteria was contained in the solution except the alcohol itself, the other constituents being only 0.5% urea, 0.1% potassium phosphate, and 0.01% magnesium sulphate. The infection was made from putrid meat and the flask left to stand at the ordinary temperature. The results were as follows :—

0.1% allylic alcohol : after 9 days, slightly turbid ; a few bacteria.
0.5% „ „ : after 24 days, quite clear, no bacteria.
1% amylic „ : after 14 days, no bacteria.
10% ethylic „ : after 10 days, slight turbidity ; small oval *yeast cells*[1] were seen but *no* bacteria.
10% methylic „ : after 5 days, slight turbidity ; small oval shaped bacteria only were seen.[2]

In a further series of experiments I compared the nutritive effect of the nine alcohols in dilutions of 0.1% with only the addition of 0.5% ammonium phosphate, 0.1% monopotassium phosphate, and 0.01% magnesium sulphate. These solutions were all infected from the same source, viz : a Pasteur's solution that had been exposed in

1. Compare O Lœw, *Natürl. System der Giftwirkungen*, S. 26.
2. According to R. Brown (*Chem. Soc. Jour.*, 1886.) *Bacterium aceti* utilises methylic but not amylic alcohol.

the open air and contained bacteria, yeast cells, and mould fungi. The results are as follows :—

Methylic	alcohol :	some development observed in one day, but increase moderate even after 21 days ; small and large yeast cells, and bacilli observed.
Ethylic	,,	like the preceding, but the increase greater ; besides yeast cells and bacilli some mucor-like mycelium was present.
Propylic	,,	like the preceding, but a mould fungus with white spores was observed in this case.
Isopropylic	,,	like the preceding (very little mycelium but numerous bacilli) ; no spore-bearing mould fungus.
Butylic	,,	after 2 days turbidity had set in, but the increase was very small even in 21 days ; bacilli and a trace of mycelium.
Isobutylic	,,	the quantity of fungi seemed a little larger than in the previous case.
Tertiary butylic	,,	the fungoid growth was here much larger than in the last two cases, many bacilli, small yeast cells, and *mycoderma*-like yeast, but no mycelium observed.
Amylic	,,	after 2 days only very little development, but considerable in 21 days ; here more mycelium was observed than in any of the other cases.
Allylic	,,	after 21 days, the liquid was perfectly clear and free from fungi.[1]

1. Parhaps a development might have been noticed if the ordinary phosphate had been used here instead of the monophosphate.

These observations of the action of the alcohols upon microbes lead to the conclusion that the common microbes of putrefaction are killed by 0.5% allylic, 2% amylic, or 20% methylic or ethylic alcohol (but probably not the spores[1]), if these substances are permitted to act for 24 hours in absence of any nourishing materials; further, that in high dilution the higher alcohols are better food for the microbes than methylic alcohol, but as the dilution lessens, methylic alcohol proves a better nutrient than the higher alcohol, the latter then showing poisonous action.

Furthermore, all my experiments on different organisms go to prove that ethylic alcohol is a stronger poison than methylic alcohol, although Dujardin-Beaumetz and Andigé (*l. c.*) have asserted the contrary, at least for higher animals. My results, moreover, agree well with those of Gibbs and Reichert (*l. c.*), who compared the action of these alcohols upon the higher animals. It is further to be concluded that the normal and iso-propylic alcohols behave nearly alike, and that of the three butylic alcohols the normal is the most poisonous, and the tertiary the least. Allylic alcohol is not only much more poisonous than the corresponding propylic alcohol but also more so than amylic alcohol. The toxic action of the allylic alcohol is evidently somewhat different from that of the saturated alcohols, whose toxical power generally increases with the number of carbon atoms in their molecule. According to Meissner[2] allylic alcohol acts 50 times more strongly than propylic. According to

1. *Koch* has found that even a much higher concentration would not kill certain spores.

2. Meissner observes: "Allylic alcohol damages the circulatory system, enlarging the blood vessels and paralysing heart-action. Allylic alcohol has *none* of the *narcotic* action of the alcohols in the saturated series.———Other important differences between allylic and other alcohols are that, when inhaled, it attacks the mucous membrane, causes great loss of protein matters, and acts *50 times stronger* than propylic alcohol." (Chem. Centralb. 1891, II 715).

my observations, however, the difference is much greater. Allylic alcohol seems to attack protoplasm directly, by chemical affinities arising from the double linking of two carbon atoms, while the saturated alcohols act merely catalytically by transferring certain kinds of motions.[1]

Living protoplasm seems a very delicate indicator of differences in chemical constitution, and when we consider how indifferent dead protoplasm is towards most of those poisons that react easily upon living protoplasm, we cannot doubt for a moment but that a great chemical change must have taken place at the moment of death in the proteids of the living protoplasm.

In conclusion, I tender hearty thanks to Prof. Dr. O. Loew for the interest he has taken in my investigation.

1. Therefore those alcohols have here been compared only in equal weights and not in equivalent quantities.