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Dietary utilization of aliphatic alcohols by *Drosophila*

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Summary. Metabolic utilization was measured by the increase in life duration of adults receiving a low concentration of alcohol as the sole energy source. Flies were able to use primary alcohols as food in the following order: butanol > ethanol > propanol. Methanol and secondary alcohols were not metabolized. These results cannot be explained by considering only the specific activity of alcohol dehydrogenase upon these substrates.

Adaptation to resources containing alcohol is a well established property of *Drosophila* species living in wine cellars[2-4]. This adaptation corresponds to a capacity to tolerate high amounts of ethanol[4,5] and also to use small amounts as a food for energy production[6-8]. For these 2 different physiological traits, alcohol dehydrogenase (ADH) plays a key role since ADH negative mutants are both sensitive to ethanol and unable to metabolize it[9,10].

The enzymatic specificity of ADH does not, however, correlate with its physiological role in the live fly. ADH is known to be inactive on methanol, slightly active on ethanol, more active on primary propanol and butanol and still more on secondary propanol and butanol[11,12]. In the living fly[9] ADH is unable to detoxify methanol, is very efficient in promoting ethanol tolerance, is poorly active in detoxifying n-propanol and n-butanol and is still less efficient with secondary alcohols.

Recently, analysis of strains selected for ethanol tolerance showed that the capacity for metabolizing small quantities of alcohol was not related to the detoxification ability[13]. This possible genetic independence of the 2 traits led us to study the relationship between the nutritive value of various alcohols and their toxicity: our results demonstrate an absence of direct correlation: for example n-butanol, which is highly toxic, is better used as food than ethanol when in low concentration.

Experiments were made on a French strain (Colmar) of *Drosophila melanogaster*, homozygous for the F (fast) allele of ADH. Groups of 10 newly emerged adults were placed in air tight vials containing a small concentration of alcohol in water. Dead flies were recorded twice a day and average longevity calculated. As in previous studies with ethanol[5,6] the metabolic utilization was estimated by increase in life duration compared to control flies receiving only water.

Results obtained with ethanol, primary and secondary propanol and butanol are given in figure 1. In the different experiments survival of control flies ranged between 60 and 70 h. With the secondary alcohols, no gain in life duration was observed and a decrease, due to a toxic effect, occurred with concentrations higher than 1%; *Drosophila* adults cannot use these alcohols as an energy source. By contrast, a significant increase in longevity was observed with the 3 primary alcohols. All the curves have the same general shape: first, life duration increases with concentration, then it decreases when the toxic effects of higher levels overcome the beneficial ones. The rapidity of the decrease is proportional to the toxicity, and we see that ethanol is the least toxic and butanol the most toxic alcohol, thus confirming previous results[9].

The nutritive values of the various alcohols can be compared by considering longevities obtained with a small, non-toxic concentration. For example, with 1% alcohol, life extension is greatest with butanol (the most toxic at higher concentrations) and shortest with propanol.

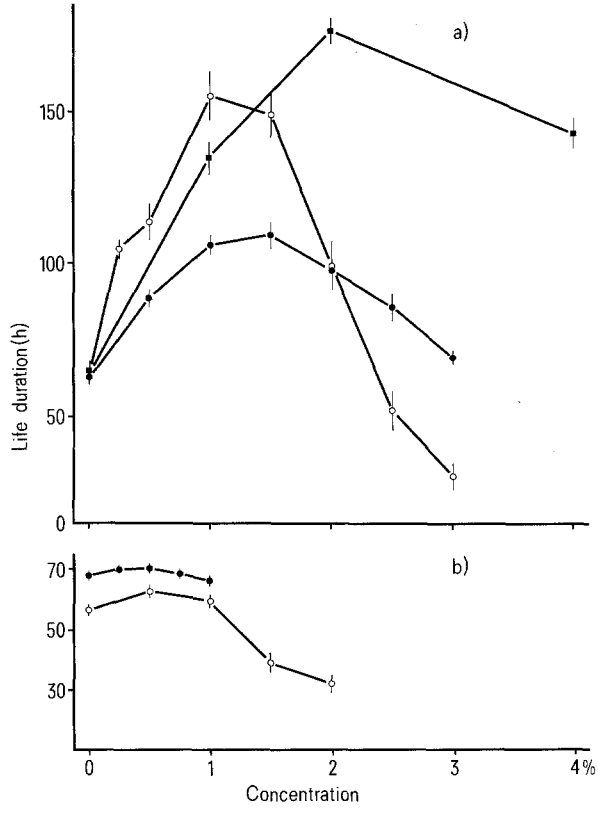

Fig. 1. Influence of alcohol concentration (% by volume) upon life duration of *Drosophila* adults. *A* Primary alcohols: ■, ethanol; ●, propanol; ○, butanol. *B* Secondary alcohols: ●, isopropanol; ○, isobutanol.

Finding that butanol surpasses ethanol was an unexpected result which needed confirmation. 2 other experiments were done using a concentration of 1% for all the alcohols studied, methanol being added to the comparison. Results (figure 2) show that methanol was not used as a food and also confirm the data of figure 1: ethanol, n-propanol and n-butanol can all be used as an energy source, butanol being the best source.

Non-utilization of methanol can be explained by the inactivity of *Drosophila* ADH on this substrate[11,12]. But another explanation is needed for secondary alcohols which, although not used, are the most readily transformed enzyme substrates. In the case of ethanol, it is generally accepted that ADH produces acetaldehyde[9,14] which is then transformed into acetate before being used in the Krebs cycle.

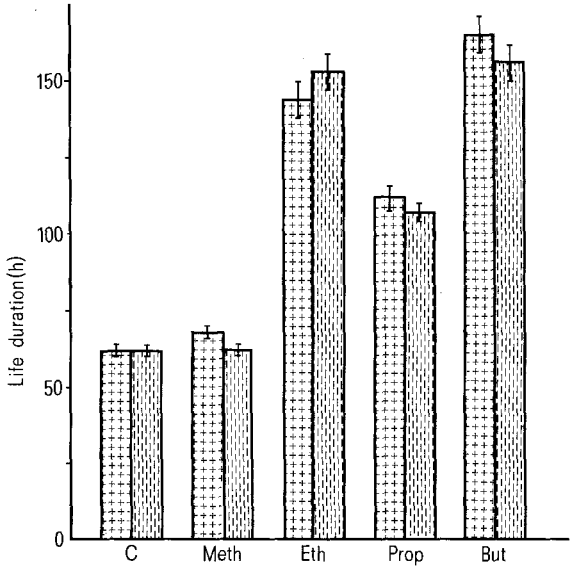

Fig. 2. Life duration of *Drosophila* adults observed with a concentration of 1% of different primary alcohols. C, controls, receiving only water; Meth, methanol; Eth, ethanol; Prop, propanol; But, butanol. For each treatment, results of 2 different experiments are given (confidence limits are shown; the number of individuals in each case was about 100).

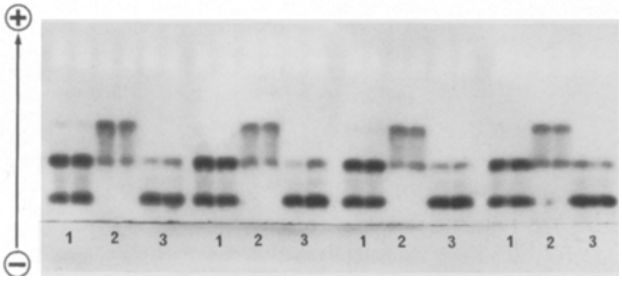

Fig. 3. Electrophoretic mobility, after starch gel electrophoresis of Adh^F homozygous flies. *1* Control, untreated flies; *2* flies treated for 24 h with 1% isopropanol; *3* flies treated for 24 h with 1% n-propanol. With isopropanol only, the modification of the electrophoretic mobility indicates acetone production (see Papel et al.[16]). The experience was repeated 4 times.

With secondary alcohols, ADH is likely to produce ketones which are not further metabolized[15]. Both acetaldehyde and acetone are highly toxic products with lethal concentrations of about 0.5%[10,16]. Preliminary experiments showed, however, that acetaldehyde has a real nutritive value, and increased the life duration of adults, while acetone did not[16]. Recently a specific poisoning of *Drosophila* ADH by acetone, which decreases its activity and modifies its electrophoretic mobility, has been described[16]. Flies were treated for a day with 0.5% of either n-propanol or isopropanol and then submitted to starch gel electrophoresis. Results (figure 3) show a modification of the electrophoretic migration of the enzyme after treatment with isopropanol, thus strongly suggesting the production of acetone with this substrate.

Primary propanol and butanol are probably transformed into aldehydes and then into propionate and butyrate. The higher nutritive value of butanol may correspond to a better capacity for metabolizing butyrate but further studies are needed.

All these data should be regarded from an evolutionary point of view. The high activity of ADH on secondary alcohols has no evident adaptive significance since these alcohols are transformed into more toxic products which are, moreover, enzyme inhibitors. Authors who, like Van Delden et al.[17], have tried to demonstrate selective effects at the *Adh* locus and have experimented with various alcohols should take our results into consideration. In nature, yeast fermentation produces mainly ethanol which, depending on concentration, can either be toxic or a nutrient. All the other alcohols are produced in very small amount so that no special detoxifying capability is needed. Adaptation to these alcohols could exist for a more efficient exploitation of available resources. We are, however, reluctant to interpret the efficient metabolization (at 1%) of n-butanol as a consequence of natural selection. Information on the quantity of the various alcohols in natural breeding sites should help to resolve the n-butanol problem.

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