concentrations (0.003 μg/g dry weight rotifer) in *B. calyciflorus* (run 1) following ingestion of plant material, but at no other time in this or subsequent runs of *B. calyciflorus* or *A. sieboldi*. Chlorophyllide b was not detected in any experiments. Chlorophyllides are degradation products formed by the action of the plant enzyme chlorophyllase and therefore probably do not represent a major degradation pathway in zooplankton. Pheophytin a and b concentrations in *B. calyciflorus* were greatest at the 3 h sampling point, decreased rapidly after 24 h, and were present in negligible or non-detectable amounts after 72 h. These observed chlorophyll degradation pigments are known to be produced in the digestive tract of other zooplankton rapidly after ingestion of viable chlorophyll[10,11]. In *A. sieboldi* pheophorbides a and b were still at high levels after 20.5 h.

The curves presented in the figure for the second experiment with *B. calyciflorus* show that chlorophyll a and its degradation products are conserved in the rotifer body during the first 24 h and subsequently decrease with time. After 24 h the sum of chlorophyll a + pheophytin a + pheophorbide a concentrations was 94.7% of the chlorophyll a and pheophytin a concentration at the beginning of the experiment. At 72 h the degradation products represent only 11% of the initial chlorophy l a + pheophytin a present initially. A similar pattern may account for chlorophyll b but it is occasionally undetected owing to the very small concentrations of chlorophyll b and pheophytin b relative to chlorophyll a. The high concentrations of pheophytin a or b relative to chlorophyll a or b present at the beginning of an experiment is probably due to rapid degradation of the chlorophyll molecule upon contact with the probably acidic rotifer gut.

The retention of chlorophyll degradation products for up to 72 h, following transfer to a chlorophyll-free diet, suggests uptake of chlorophyll a and b or their degradation products. Evidence for the uptake of chlorophyll in rotifers[12,13] and other invertebrates[14,15], and its possible subsequent use in pigmentation[16] and biosynthesis is rare but not unknown.

The most important information in the present study concerns the relative uptake and retention of tocopherol as compared with chlorophyll. The measured molecular tocopherol : chlorophyll a ratio in *E. gracilis* (1:41.6) indicates the low concentration of tocopherol relative to chlorophyll a molecules; the tocopherol : chlorophyll b ratio is 1:7.3. Both measurements on *B. calyciflorus* feeding on *Euglena* show high molecular tocopherol : chlorophyll a ratios (2.4:1 and 2.5:1) and tocopherol : chlorophyll b ratios (43.5:1 and 72.0:1). In *A. sieboldi* the initial molecular tocopherol : chlorophyll a ratio was 4.4:1 and the tocopherol : chlorophyll b value was 154.3:1. The small discrepancy in tocopherol : chlorophyll a or b ratios between *A. sieboldi* and *B. calyciflorus* may be due to their different modes of feeding. *Brachionus* is a herbivore and ingested algae and algal fragments directly whereas *Asplanchna* is a carnivore and obtained algal products secondarily by feeding on *Paramecium*. The *Paramecium* diet of *A. sieboldi* probably contained a higher tocopherol : chlorophyll pigment content than the algae eaten directly by *B. calyciflorus* owing to minimal tocopherol degradation and high chlorophyll pigment degradation and loss during intracellular digestion. Clearly then, the rotifers are assimilating tocopherol in preference to the more abundant chlorophyll a and b molecules.

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The Relationship of the Vertical Distribution of Japanese Conifers to the Maltol Contents in their Leaves

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Summary. Of 35 species of coniferous trees in Japan, only 4 species, found at subalpine zone, contain maltol in their leaves. Their leaves are also characteristic for their high content of sugar which protects the plants against freezing. It is interesting from the viewpoint of plant ecology that only the above 4 species contain high amounts of sugars and maltol in their leaves because maltol is biosynthesized from sugars.

There are 35 species of coniferous trees in Japan, a relatively large number considering Japan's geographical position. Of these 35 species, only 4 contain maltol (2-methyl-3-hydroxypyran-4-one) in their leaves. They are *Abies mariesii* (Japanese name, Oushirabiso), *A. veitchii* (Shirabiso), *A. homolepis* (Urajiromomi) and *Tsuga diversifolia* (Kometsuga). It is characteristic of these 4 species that they are found only at elevations between 1500 and 2900 m, that is to say that they are common to the subalpine zone alone and are not found lower (Figure 1). Japan's other 31 coniferous species (19 species of the Pinaceae family, 2 species of the Taxodiaceae family, and 10 species of the Cupressaceae family) are generally found at lower levels, ranging from sea level to 2000 m. In the lower subalpine zone (1500–2000 m), the vertical distribution of the above-mentioned 4 conifers and the rest of Japan's coniferous species overlap. It should be noted, however, that while *A. mariesii* and *T. diversifolia* resemble *Abies firma* (Momi) and *Tsuga sieboldii* (Tsuga) respectively, only the former leaves bear rich maltol (0.5–3.4%). The vertical distribution of *Pinus pumila* (Haimatsu) is wide (Figure 2). In Honshu it can be found at elevations as high as 3200 m, and in Hokkaido it is common from sea level to the critical forest limit, which, owing to the latitude, is at about 2300 m. Maltol is not found from the leaves of *P. pumila*. Maltol was first isolated from *Larix decidua*[1], a species common to Europe. It is interesting to note that maltol is not found in *L. leptolepis*, the Japanese variety of *L. decidua*, which is found below the subalpine zone.

In contrast, the 4 conifers that have high maltol contents have a narrow vertical distribution. They are found only in the subalpine zone as already stated. They are the oldest plants that were already distributed and flourished prior to the great ice age in Honshu. Our knowledge of geographic history teaches us that there was a familiar exchange between the conifers and circumpolar plants of the old continent before Honshu was separated from the continent[2].

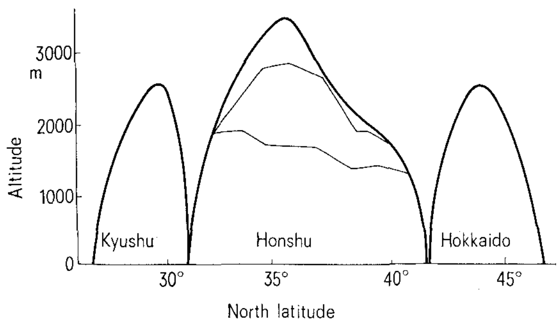

Fig. 1. The vertical distribution of *Abies mariesii*, *A. veitchii*, *A. homolepsis* and *Tsuga diversifolia* (Y. Hayashi).

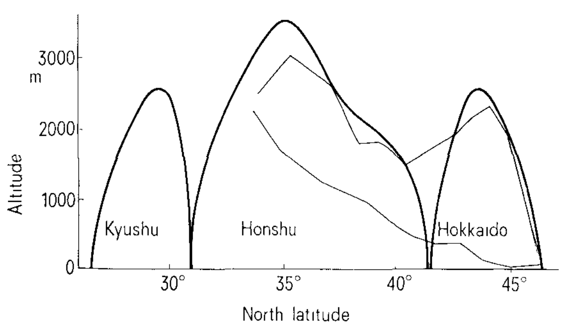

Fig. 2. The vertical distribution of *Pinus pumila* (Y. Hayashi).

The leaves of the 4 species of conifers that have high maltol contents are also marked by high sugar contents, which protect them against freezing[3]. In addition, the lower epidermises of these leaves are coated with a white waxy layer in which maltol is abundant. Maltol has sweet odour that is often used as a flavour enhancer and, as a result, these 4 species have a characteristic sweet smell.

Because maltol can be produced simply[4] by heating sugar, by fermenting malt, or by cooking food, it is believed that maltol may be biosynthesized from sugars in plants. At any rate, there seems to be a definite connection between the presence of sugars and maltol in the leaves of the 4 species of conifer. In particular, it is interesting from the viewpoint of plant ecology that high amounts of maltol and sugars are contained in leaves of these conifers that ranged only in a subalpine zone in Japan.

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The Generation of Toxic Activity from *Trypanosoma congolense*[1]

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Summary. Trypanosoma congolense organisms, on incubation at 20 °C for $9^1/_2$ h, were found to generate phospholipase-like activity which was capable of mediating lysis of both nucleated cells and erythrocytes as well as an acute inflammatory response on intradermal inoculation.

Although it was suggested long ago that salivarian trypanosomes may produce 'toxins'[2,3], evidence for the existence of these has generally been fragmentary and inconclusive. The toxic activities which have been demonstrated in trypanosome preparations from various species include, the capacity to cause acute inflammation on intradermal inoculation[4] and around intraperitoneal diffusion chambers[5], an ability to induce mild narcosis in mice[6], and the production of hemolytic factors[7,8]. Nevertheless, none of these have been unequivocally established as playing a significant role in disease due to the African trypanosomes.

In this communication we report on the generation of toxic activity from *T. congolense* (Strain TREU 112) which we consider to be of potential significance in the pathogenesis of disease caused by this organism. During the course of experiments on dermal hypersensitivity to *T. congolense* in rabbits it was observed that a single intradermal injection of 0.05 ml of a 1.0% v/v suspension

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