non-O-glycosylated basic flavone isovitexin itself causes this striking morphological effect.

A microscope with quartz optics in combination with a monochromator enabled us to locate flavones. These studies showed that the isovitexin (glycosides) are mainly present in the vacuoles of the upper epidermis cells (unpublished results). Scanning elctron microscopic studies of petal surfaces from individuals with and without a glycosylated derivative of isovitexin revealed little difference in the lower epidermis; in both cases the elongated cells fit as in a jigsaw puzzle and are arranged in regular arrays. The upper epidermis of petals from normal plants also shows a regular appearance and arrangement (fig. 3a), but the cells in the upper epidermis of petals of the aberrant plants we found to be much less regular. Swollen cells are succeeded by groups of flat cells, and some of the swollen cells appear to have burst, their rims still protruding (fig. 3b and c). The different types of aberrant cells are not evenly distributed over the upper epidermis. In the basal part of the petal swollen cells are the most common cell type; burst cells are most common in the central part. Flat cells are especially numerous towards the apex.

It appears then that isovitexin has some toxic effect on the cells of the upper epidermis. This toxication leads to swelling of the cells at the base of the petal; in the middle of the petal it leads to collapse, whereas the flat cells at the top either are the debris of the burst cells, or are cells which have failed to develop.

The question remains why in *Silene pratensis* the accumulation of the flavone isovitexin in the vacuole causes this morphological effect. It is possible that the glycosylation of isovitexin prevents leakage across the tonoplast and thus prevents its interaction with the cell components of the cytoplasm. In vitro, flavonoids can influence many processes at very low concentrations, varying from indole acetic acid catabolism and hence hormone balance[12-14], to DNA replication[15-17] and oxidative- and photo-phosphorylation[14,18,19,21]. Among others Stenlid[14] showed that flavones can act as potent inhibitors of ATP synthesis in mitochondria, comparable to the classical uncoupler dinitrophenol. Flavone aglycones are in this respect many times more active than their glycosylated derivatives[13,14,18]. We hypothesize that free isovitexin interferes with the energy supply of the upper epidermal cells, which therefore have difficulties with the maintenance of turgor, leading to swelling and ultimately to bursting. The protruding debris of these cells may be rubbed off and thus they give the impression of flat cells. Finally, the damage done to the upper epidermis leads to the curling-up of the petals.

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Effect in heavy meromyosin on conformation of F-actin

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Summary. Cooperative conformational changes of F-actin induced by heavy meromyosin (HMM) binding (in the absence of troponin and tropomyosin) were found by the method of polarized UV-fluorescence microscopy.

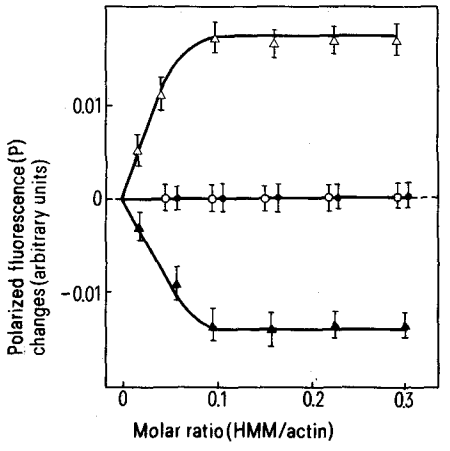
Binding of myosin to F-actin is known to be an important moment in the generation of tension in a muscle fiber. However, the conformational changes of F-actin during its interaction with myosin are still insufficiently studied. In the present study, the changes of the state of F-actin at HMM binding have been explored by the method of polarized UV-fluorescence microscopy.

Materials and methods. The study was carried out on glycerinated ghost single fibers of rabbit muscle[1,2]. Such fibers were free of myosin, troponin and tropomyosin[2,3] and contained more than 80% of actin[3]. In some experiments, the fibers were treated with 10% glutaraldehyde for 1 min. HMM was prepared by tryptic digestion of rabbit skeletal myosin using the method of Szent-Györgyi[4]. The Ca^{2+}-ATPase activity of HMM was 0.95 μmoles P_i/min/mg when measured at low salt concentration, pH 7.5 at 25 °C. F-actin was decorated with HMM by incubation of a ghost single fiber in a solution containing 5 mg/ml HMM, 60 mM KCl, 1 mM $MgCl_2$, 50 mM Tris-HCl, pH 7.0. The intensity of fluorescence (I_m) and the degree of polarization of tryptophane fluorescence (P) was measured by polarized microfluorimetry[5]. P was registered at fiber orientations both parallel ($P_\parallel$) and perpendicular ($P_\perp$) to the plane of the exciting light. All measurements were

performed in a solution containing 100 mM KCl, 2 mM $MgCl_2$, 67 mM phosphate buffer, pH 7.0.

Results and discussion. It has already been shown that after extraction of myosin, troponin and tropomyosin from a ghost single fiber there remain tryptophane residues of F-actin, their dipoles of emission being oriented preferably perpendicular to the fiber axis[1-3], i.e. anisotropically. On the other hand, tryptophane residues of proteins other than F-actin are arranged practically isotropically. Furthermore, their contribution to the general emission of a fiber is much lower than that of F-actin, therefore $P_\perp > P_\parallel$[1,3]. $P_\perp$ and $P_\parallel$ depend on the optical properties of F-actin tryptophane residues, the latter being defined by the structural parameters of the actin helix in the thin filaments. Changes in $P_\perp$ and $P_\parallel$ are therefore sensitive to the conformational changes of the protein[1].

HMM binding to F-actin decreases the anisotropism of tryptophane fluorescence of a ghost single fiber, resulting in $P_\perp$ decrease and $P_\parallel$ increase (fig.). Since the anisotropism is not sensitive to orientation of myosin heads in fibers[6], the changes of $P_\perp$ and $P_\parallel$ indicate conformational changes of F-actin induced by HMM binding.



Dependence of changes of polarized fluorescence (P) on the molar ratio of HMM to actin monomers (HMM/actin) of ghost single fibers (△—△, ▲—▲) and fibers treated with glutaraldehyde (○—○, ●—●) after addition of 5 mg of HMM/ml. △—△, ○—○; ▲—▲, ●—● – fibers at parallel and perpendicular orientation to the plane of the exciting light, respectively. Measurements were carried out in a solution containing 100 mM KCl, 2 mM $MgCl_2$, 67 mM phophate buffer, pH 7.0. Each point is the average of 50 measurements obtained in 10 fibers. Vertical bars show SD.

It should be noted that the changes of fluorescence anisotropism are only observed if F-actin is capable of changing its conformation. Thus, glutaraldehyde-treated F-actin binds HMM, the interaction being functionally effective as measured by ATPase activities, but, the conformation of F-actin remains unchanged due to fixation[7], therefore $P_\perp$ and $P_\parallel$ are constant (fig.).

HMM binding to F-actin induces an increase of fluorescence intensity. Since after decoration I_m increases due to HMM tryptophane residues, it is possible to estimate the molar ratio of HMM to actin. For example, an increase of I_m by 25% corresponds approximately to a molar ratio of 0.1, since according to the data of Yanagida and Oosawa[3] an increase of fluorescence intensity by 80% reflects a molar ratio of 0.3. The decrease of tryptophane fluorescence anisotropism was not directly proportional to the amount of bound HMM (fig.). The binding of 1 single myosin head to 10 actin protomers is enough to induce maximal changes of tryptophane fluorescence anisotropism of F-actin in thin filaments. This phenomenon suggests a conformational change in neighboring actin molecules which is induced co-operatively by the conformational change in the actin molecule on the binding of HMM.

From the use of polarized fluorescence analysis[3,8], and having made a mathematical model of the data obtained, it can be assumed that HMM binding induces cooperative changes of the orientation of protomers in F-actin as well as an increase in the flexibility of the thin filament. The results show a certain amount of agreement with those which have been obtained by other methods[3,9,10]. Such structural changes of F-actin might be an important factor in the interaction of actin with myosin during muscular contraction.

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Intra-aortic prostaglandin E_1 infusion in maturation of neuroblastoma

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Summary. A $7^2/_3$-year-old boy with unresectable abdominal neuroblastoma received an intra-aortic prostaglandin E_1 infusion (0.4–0.5 ng/kg/min) over a total period of 6 months, as well as systemic papaverine and multiagent chemotherapy. At second-look surgery 9 months later, tumors grossly appearing to be ganglioneuromatous were subtotally resected. Histology revealed the evidence of neuroblastoma maturation.

The prognosis of disseminated neuroblastoma remains poor[1]. In vitro and in vivo studies demonstrate that neuroblastoma differentiates and maturates into ganglion-like cells under the effect of adenosine-3′,5′-monophosphate (cAMP)[2-4]. Clinical application of these findings is worth attempting in order to induce tumor maturation and im-