

During the growth of the nucleus, the nucleolar material is gradually fragmented into smaller and smaller granules and aggregates. In the advanced previtellogenetic stages, nearly the entire nucleus is evenly filled up with an almost homogenous substance rich in RNA (Figures 3 and 4), which is composed of very small granular nucleoli, similarly as it is in the nuclei of the growing oocytes of Dytiscidae in which the nucleolar extra DNA occurs⁵. Only a relatively small area of the nucleus remains free of the nucleolar material. In this area all the oocyte chromosomes are accumulated forming a compact karyosphere. This indicates that the behaviour of chromosomes in the oogenesis of Gyridae does not essentially differ from that of the chromosomes in nuclei of the growing oocytes in the majority of insects with polytrophic ovaries.

The very significant increase in the volume and surface of the oocyte nuclei in *Gyrinus* and the abundance of the nucleolar material on the one hand, and the condensed state of the oocyte chromosomes on the other hand, seems to indicate a considerable transcriptional activity of the extra DNA contained in those nuclei. The autoradiographic examination carried out with the use of ³H-uridine has fully confirmed this supposition. The results obtained have proved that during the whole period of previtellogenesis the oocyte nuclei are very active in RNA synthesis. The intensity of this synthesis seems to be of the same order as the intensity of RNA synthesis in the nurse cell nuclei (Figure 5). The labelling of the oocyte nuclei resulting from the specific incorporation of ³H-uridine into newly synthesized RNA is evenly distributed all over them, in line with the distribution of the nucleolar material (Figure 6).

We did not succeed, so far, in demonstrating that the nucleolar RNA synthesized in the oocyte nuclei of *Gyrinus* is next transported through the nuclear membrane into the ooplasm. The fact that such a process, most probably very intensive, actually takes place throughout the whole previtellogenetic growth period of oogenesis, seems to be confirmed by the great RNA concentration in a thick layer of ooplasm adjacent to the nuclear membrane (Figures 3 and 4). The degree of basophilia of that layer of ooplasm begins to decrease only at the end of previtellogenesis, that is at the stage directly preceding the degeneration of nurse cells. It also seems that the contribution of nurse cells in supplying RNA to the ooplasm of growing oocyte is minor, at least in terms of quantity, in comparison to the role played by the oocyte nucleus in that process. The presented data are preliminary results of current investigations.

Zusammenfassung. Autoradiographische Untersuchungen an den Oozytenkernen des Taumelkäfers *Gyrinus natator* zeigen eine hohe RNA-Syntheseaktivität, kurz nachdem sich der extrachromosomale Chromatinkörper im Karyoplasma aufgelockert hat. Da die Oozytenchromosomen gleichzeitig in der Karyosphäre zusammengeballt sind, wird angenommen, dass die Transkriptionsaktivität an der extrachromosomalen DNA abläuft.

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Active Components of *Sargassum tortile* Effecting the Settlement of Swimming Larvae of *Coryne Uchidai*

Hydrozoa of Coelenterata are observed very often attached to various algae. This associative relationship between epiphytic hydrozoa and algae is formed by the sequence of settlement of hydrozoan larvae onto the associated algal thallus and then the growth of the hydrozoan colony thereon. It has been noticed biologically that most of epiphytic hydroids have their own preferred alga and that this particular association is established by

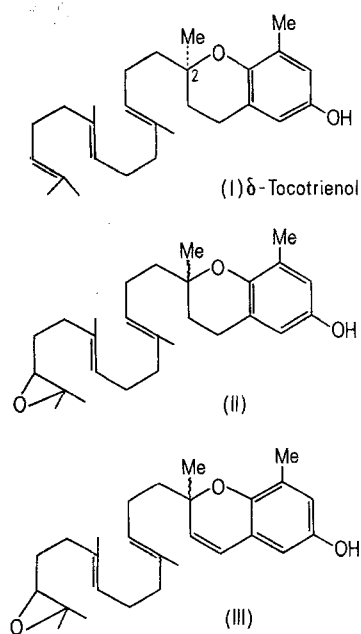
the algal preference of the settling larvae. These observations suggest that an alga, to which the swimming larvae prefer to settle, might produce some specific chemical compound which induces the settling of swimming hydrozoan larvae.

It was found recently that the settling of the swimming larvae of *Coryne Uchidai*, a kind of hydrozoa, was clearly induced by adding the juice of *Sargassum tortile* (Japanese

Bioassay of synthetic I, II and III toward the larvae of *Coryne Uchidai*

Time (h)	12 ~ 24					48					72							
	m	cl	s	a	b	p	m	cl	s	a	b	p	m	cl	s	a	b	p
δ-Tocotrienol (I) ^b		6	2		2		7				1	2				7 ^d		3
Control ^c	2	8					10							10				
Epoxide (II) ^e			3	7					5			5						5
Epoxide (II) ^f		3	4	3			2		5	1	2							7
Control ^c		5	5				5	5						2	3	5		3
Dehydro epoxide (III) ^g		5	2	3					1	4		5					4	6
Control ^h	4	2	2				4	2	4				3	1	5	1		

^aAbbreviation; m, swimming; cl, crawling; s, settling; a, attaching; b, formation of tentacle bud; and p, formation of polyps. ^bOne drop (0.05 ml) of ethanol solution containing 15 mg of DL-(I) in 1 ml of ethanol was added to 10 larvae in 20 ml of sea water. The values show the number of larvae in different stages. ^cOne drop of ethanol containing no material was added under same conditions. ^dAll of the 7 larvae died accompanying cytolysis. ^eOne drop of ethanol solution containing 30 mg of II was added to 10 larvae in 20 ml of sea water. ^fA quarter of 1 drop of the above original solution was used. ^gOne drop of ethanol solution containing 30 mg of epoxide (III) in 1 ml of EtOH was dropped on filter paper and the solvent was evaporated. The filter paper was put in 20 ml of sea water containing 10 larvae. ^hFilter paper without material was put in sea water.



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References on the biological properties of several kinds of hydrozoa are cited therein.

² Ph. D. thesis of A. S. KUMANIRENG, Tohoku University (1973).

³ Details of the syntheses will be described elsewhere.

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name: Yoremoku), a kind of alga to which the larvae settle specifically¹. This finding has forced us to elucidate the active substance in the algae.

The neutral part of *n*-hexane extracts of the dried alga (3.5 kg) was fractionated by the aid of column chromatography of silica gel for bioassay, and a portion (302 mg) of the fractions showed a specific and powerful activity in favour of the settling and subsequent metamorphosis of the swimming larvae of *Coryne Uchidai*. By further purification with preparative silica gel thin layer chromatographies, the active portion was separated into 6 compounds, i.e., A (22 mg, C₂₇H₄₀O₃), B (89 mg, C₂₇H₄₀O₂), C (24 mg, C₂₇H₄₀O₃), D (11 mg, C₂₇H₃₈O₃), E (6 mg, C₂₇H₄₀O₂) and F (18 mg, C₃₀H₄₂O₂).

On the basis of chemical and physical evidence², structure of B and A was deduced as δ -tocotrienol (I) and its epoxide (II). The deduction was unequivocally confirmed by the synthesis³, in which DL-epoxide (II) was obtained by the reduction of dehydro-epoxide (III). Since both epoxides have the same R_f on TLC and (II) shows M⁺ and (M-2)⁺ in its mass spectrum, the contamination of III in A is not ruled out at present. The remaining compounds have the similar physical properties and the structural elucidation is the subject of future investigation.

It was found that our synthetic materials, especially both epoxides (II and III), are effective in the assay using swimming larvae of *Coryne Uchidai* as shown in the Table⁴.

Résumé. Le δ -tocotriénol (I) et ses dérivés (II), tirés de l'algue *Saragassum tortile*, ont été identifiés comme étant les substances favorisant la fixation spécifique sur l'algue des larves mobiles de *Coryne Uchidai*. Les produits de synthèse (I et II) possèdent la même activité.

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The Influence of Proteolytic Enzymes on the Phosphorylation of Rat Liver Histones

There is increasing evidence that cells from tissue culture or isolated cell nuclei respond to the addition of proteolytic enzymes with stimulated synthesis of DNA¹, RNA^{2,3} or cell multiplication^{4,5}. Effective agents were trypsin¹⁻³, papain^{6,7} and lysosomal preparations⁴. In addition, activated cell proliferation was observed *in vivo*, following *i.v.* or *i.p.* administration of proteases⁶⁻⁸. This paper describes an enhanced phosphorylation of certain histone fractions, caused by *i.p.* injections of low doses of trypsin and papain. The investigations were suggested by the observation that cellular proliferation is dependent on regulatory influences of nuclear proteins, and that the amount of phosphate or acetate incorporated into specific histone fractions, varies with the stage of the cell cycle⁹⁻¹¹.

The Figure demonstrates a significant stimulation of phosphate incorporation into liver histones F1, F1'' and F2b after a single injection of papain (0.5 or 3 mg/animal) or trypsin (3 or 9 mg/animal). A similar effect is observed with fraction 'c', a protein component, not belonging to the histone group¹². The stimulated phosphate uptake

can be produced also by application of Wobe-Mugos (Mucos-Emulsionsges. mbH., Grünwald/München, 0.2 and 1.2 ampoules/animal), an enzyme preparation used in

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