REVIEW

Phylogeny and evolution of chemical communication: an endocrine approach

A M Stoka

Centro Regional de Investigaciones Científicas y Tecnológicas (CRICYT) Mendoza, Argentina
Delegación Sanitaria Federal, Ministerio de Salud y Acción Social, Mendoza, Argentina
Facultad de Ciencias Médicas, Universidad Nacional de Cuyo, Mendoza, Argentina

(Requests for offprints should be addressed to A M Stoka at J B Justo 370, piso 2, dto 22, 5500 Mendoza, Argentina)

ABSTRACT

The present review assesses the phylogenetic history of information molecules (bioregulators, pheromones, hormones, neuroactive compounds), receptors, transducers, second messengers) in uni- and multicellular organisms.

Transitional stages between contemporary endocrine secretions including hormones and neuroactive materials, and primogenial exocrine

compounds (pheromones) are proposed. Several hypotheses have been developed to explain the origin and evolution of bioregulator/receptor units.

Finally, how these primordial information molecules have either been co-opted or have changed their function during the course of biological evolution is analysed.

Journal of Molecular Endocrinology (1999) **22**, 207–225

INTRODUCTION

Successful biological evolution may be considered to have, in large part, resulted from chemical communication using messengers such as food signals and toxins; these may be regarded as the primary conduits for information in the biosphere. The emergence of multicellular organisms, such as vertebrates, necessitates the differentiation and specialization of multiple chemical signalling systems organized in a network to carry the diffusible signal from one cell to the other (Fig. 1) either between or within several compartments.

The existence of an endocrine system as such conceptually demands multicellularity, but the actions of bioregulators are not restricted to the classical concept of hormones as endocrine signals. The tenor that 'compounds secreted by endocrine structures (glandular or nervous cells) and transported to other parts of the body by way of the blood-stream (or other fluids), where they evoke physiological responses' no longer applies.

Indeed, many bioregulators such as pheromones, paracrine substances, and growth factors do not

originate from defined glandular or nervous structures and are delivered to their sites of action in many ways (Fig. 1). Chemical communication is central to evolutionary history; many types of information molecules including bioregulators, receptors, transducers, effectors and second messengers are almost universally present. Uni- and multicellular organisms are likely to share biosynthetic and functional mechanisms in terms of their chemical communication.

The use of phylogenetic perspectives to reconstruct evolutionary history has become increasingly important and provides solutions to basic questions in the area of chemical communications. Bioregulators, secretory cells and target tissues have left no palaeontological record, but the evolution of chemical communication can be extrapolated from comparative analyses of extant organisms.

In addition, the living record is of course far richer and more extensive than the fossil one, because cells today retain important information about their past in terms of amino acid sequences of their proteins and in the composition of nucleic acids (RNA, DNA).

FIGURE 1. Interactions among chemical signalling systems and their respective environments. Bioregulators travelling through internal (blood stream, haemolymph) or external (water, air) environments are called hormones (or neurotransmitters, etc.) and pheromones respectively. In some cases, a bioregulator acts simultaneously as hormone and pheromone as occurs in the interaction between host and parasite (see Interspecific Communication).

Thus, a phylogenetic analysis of the distribution, biological functions and mechanisms of action of informational molecules is a logical way to gain insight into evolutionary trends and history.

INTRASPECIFIC COMMUNICATION

The ubiquity of bioregulators throughout the biosphere suggests that these compounds predate truly hormonal roles, and chemical communication took place among unicellular organisms prior to the emergence of Metazoa.

Two primary secretory systems may be recognised depending on the routes they take following release. One type involves the synthesis and release of materials which have actions within the organism itself (contemporary endocrine system). The second type of secretion includes pheromonal systems in which the secretion leaves the organism, either as liquid or gas, to affect the function of a second organism (primordial exocrine system).

The variety and ubiquity of pheromonal molecules (cyclic nucleotide, amino acids, small peptides, large proteins, alkanes, ketones, terpenoids, steroids; Table 1, Fig. 2) renders it likely that such substances in unicellular organisms evolved into the hormones of multicellular organisms.

The earliest phylogenetic example of intraspecific communication at cellular organization level is the aggregation process (Bonner 1971, Schapp 1984) of unicellular organisms of the same species in which there is directed migration towards a region of higher concentration of pheromone (paracrine signal) (Table 1).

In addition, some pheromones are produced in the same cell in which they exert their effects, referred as to an autocrine signal; this type of self-stimulation is seen in the protozoan *Euplotes raikovi* (Vallesi *et al.* 1995). There are two kinds of pheromone–receptor interactions in this species: (i) autocrine pheromone receptors (cell division) and (ii) paracrine pheromone receptors (mating behaviour). In the first type of interaction the pheromones cause mitogenic proliferation of the same cells from which they were secreted. In the second type of interaction these cells interrupt their vegetative cycle and are triggered towards mating behaviour when non-self co-specific pheromones act on paracrine pheromone receptors.

There are broad paradigms for the evolution of the chemical communication involved in these biological responses: (i) the same information molecule can develop more than one physiological function; (ii) the autocrine and paracrine functions appeared prior to the origin of endocrine function; (iii) reciprocal interaction between members of the same species began in an asocial environment (without colonies and without co-operative interactions) preceding the development of multicellular organisms.

Chemical communication ranges from prokaryotes responding to a wide variety of environmental chemical signals to the complex endocrine regulatory processes in multicellular organisms. Certainly, the ubiquity of the vertebrate information molecules (Table 2) is a product of natural selection–adaptation events.

It is, however, important to distinguish between characteristics selected for one function (adaptation process) from those originally selected for one function but subsequently employed by chance in an extension of the initial function (exaptation process) (Gould & Vrba 1982).

A particularly good example of this is seen in invertebrate and vertebrate metamorphoses, spectacular biological processes regulated by a variety of hormones (Bentley 1976, Stoka 1987). In this process, juvenile hormone (sesquiterpenoid, Insecta) and prolactin (polypeptide, Amphibia) act as juvenile agents, although they are also involved in reproductive functions (Bern & Nicoll 1969, Highnam & Hill 1977). The ancestral insects and amphibians were essentially aquatic, metamorphosis

TABLE 1. Ubiquity and chemical diversity of pheromones

Organism	Effect	Chemical diversity	Reference
Algae			
<i>Volvox carteri</i>	Cellular differentiation	Glycoprotein	Starr & Jaenicke (1974)
<i>Ectocarpus siliculosus</i>	Sexual attraction	Ectocarpin (Fig. 2)	Muller <i>et al.</i> (1971)
<i>Fucus serratus</i>	Sexual attraction	Fucoserraten (Fig. 2)	Muller & Jaenicke (1973)
Fungi			
<i>Dictyostelium lacteum</i>	Aggregation	Pteridine derivative	Van Haastert <i>et al.</i> (1982)
<i>Polysphondylium violaceum</i>	Aggregation	Glorin (Fig. 2)	Shimomura <i>et al.</i> (1982)
<i>Allomyces sp</i>	Sexual attraction	Sirenin (Fig. 2)	Machlis <i>et al.</i> (1968)
<i>Achlya bisexualis</i>	Cellular differentiation and sexual attraction	Antheridiol (Fig. 2)	Barksdale (1967)
<i>Achlya bisexualis</i>	Idem	Oogoniol (Fig. 2)	McMorris <i>et al.</i> (1975)
<i>Sacharomyces cerevisiae</i>	Idem	Alpha and a factors (Fig. 2)	Wilkinson & Pringle (1974); Duntze <i>et al.</i> (1970)
<i>Mucor mucedo</i>	Cellular differentiation	Trisporic acid (Fig. 2)	Meselson & Radding (1975)
Protozoans			
<i>Euplotes rathkovi</i>	Cell division and mating behaviour	Polypeptides	Vallesi <i>et al.</i> (1995)
Arthropods			
<i>Bombyx mori</i> (Insects)	Sexual attraction	Bombycol (Fig. 2)	Butenandt (1963)
<i>Apis mellifera</i> (Insects)	Sexual attraction	9-oxo-decenoic acid (Fig. 2)	Butler (1967)
<i>Balanus balanoides</i> (Crustaceans)	Aggregation	Protein	Crisp & Meadows (1962)
Fishes			
<i>Carassius auratus</i> (goldfish)	Sexual behaviour	17,20 β -dihydroxy-4-pregnen-3-one	Dulka <i>et al.</i> (1987)
<i>Fugu niphobles</i>	Sexual attraction	Tetrodotoxin	Matsumara (1995)
Amphibians			
<i>Cynops pyrrhogaster</i>	Sexual attraction	Sodefrin (Fig. 2)	Kikuyama <i>et al.</i> (1995)
Reptiles			
<i>Thamnophis sirtalis parietalis</i>	Sexual attraction	Long chain C ₂₉ -C ₃₇ unsaturated methyl ketones	Mason <i>et al.</i> (1989)
Mammals			
<i>Sus scrofa</i>	Sexual attraction	Androgen steroids	Melrose <i>et al.</i> (1971)
<i>Homo sapiens</i>	Sexual behaviour	Androstenol (steroid)	Gustavson <i>et al.</i> (1987)

FIGURE 2. Lipid-type and peptide-type structures of exocrine bioregulators (pheromones). 1, Antheridiol; 2, oogoniol; 3, bombycol; 4, fucoserraten; 5, sirenin; 6, ectocarpin; 7, trisporic acid; 8, 9-oxo-decenoic acid; 9, glorin; 10, sodefrin; 11, alpha-factor; 12, a-factor. Alpha-factor from *Saccharomyces cerevisiae* has structural homology and functional analogy with a vertebrate-type hormone (see Table 2).

being an adaptation process to permit aquatic and terrestrial life styles (Toms 1984).

Can the primordial functions of juvenile hormone and prolactin be identified?

Insect metamorphosis is linked to sexual maturation and the primordial function of juvenile hormone was probably to regulate the reproductive cycle of the ametabolous ancestors (Apterygota) of winged insects. The most primitive contemporary insects, Apterygota, display virtually no metamorphosis and juvenile hormone acts only on reproductive tissues.

In contrast, prolactin may be seen as an osmoregulatory hormone in fish. Thus, the morphogenetic and regulatory effects of the juvenile hormone and the later physiological attributes of prolactin (milk secretion in mammals, crop-milk production in birds, water-drive behaviour in amphibians) are secondarily evolved, when juvenile hormone and prolactin were used by other tissues or species.

The acquisition of new advantageous functions by a determined bioregulator might be an opportun-

istic process in which novel structures (receptors, transducers, effectors) and new domains of an old bioregulator developed new physiological functions in other tissues and/or species. This kind of evolutionary opportunism characterizes the phylogeny of chemical communication (exaptation process).

INTERSPECIFIC COMMUNICATION

Evolutionary opportunism is commonly seen in the interactions between hosts and parasites. Examination of these relationships reveals that in some cases there are indeed effects on the hormonal regulation of development. Several studies (Table 3) described cellular receptors of host hormones in parasites which can respond differently with respect to their hosts (heterotrophic effects).

With respect to the effect of host hormones on parasites (Table 3) there is evidence for the hypothesis that some bioregulators originally served

as a defence molecule: (i) vertebrate-type steroid hormones are present in insects (Schildknecht *et al.* 1966, 1967) and steroid derivatives occur in coelenterates (Sturaro *et al.* 1982), and they act as defence mediators towards other species; (ii) sterols and steroids have antimicrobial effects (Buetow & Levedahl 1964); (iii) host hormones can inhibit the growth of parasites (Loose *et al.* 1983, Schar *et al.* 1986, Stoka 1996); (iv) several phytocompounds disrupt insect development (Stoka *et al.* 1987a).

Examples of transitional stages in the evolution of defensive and hormonal functions may also be noted: (i) a neurotoxin is a male attractant pheromone in vertebrates (Matsumara 1995); (ii) enterotoxins induce steroidogenesis in adrenocortical cells (Donta & Moon 1974); (iii) cholera toxin β -chain and vertebrate glycoprotein hormones (thyroid-stimulating hormone, luteinizing hormone, follicle-stimulating hormone) share structural similarities (Kurosky *et al.* 1977); (iv) the finding of chemical signalling systems in plants that play dual roles as growth regulators and as a defence mechanism against pathogens (Chang *et al.* 1993, Chen *et al.* 1993).

Heterotrophic effects can also be viewed as (i) an adaptive strategy of parasites that renders it advantageous to co-ordinate their physiological functions with the physiological stage of their host. For example, 20-OH-ecdysone acts as a moulting hormone in insects, but can also stimulate growth and sexual differentiation in the protozoan parasite *Trichonympha sp.* (Cleveland 1959); (ii) a mechanism for pre-existing hormones to evolve in other target tissues or species (exaptation process). In unicellular organisms vertebrate-type bioregulators affect their growth (Table 3), while in multicellular organisms they are involved in sexual activity and metamorphosis (Baulieu *et al.* 1978, Geuns 1978, Stoka 1987, Stoka *et al.* 1987b). It is likely that receptors originally triggered one physiological response after interacting with a specific bioregulator, but subsequent changes (e.g. coupling with different transducers and effectors) allowed the development of additional physiological responses. This adaptive acquisition, with an old bioregulator playing a new function, is perhaps the most important consequence of transitional stages of change.

EVOLUTIONARY ASPECTS OF EXOCRINE AND ENDOCRINE SYSTEMS

Transitional stages are, phylogenetically, key points during the divergent evolution of uni- and multicellular organisms. In an evolutionary sense primitive pheromones are a transitional stage

between the most primitive (food signals, toxins) and the most advanced (hormones, neuroactive compounds) chemical signals.

Current data are in agreement with the hypothesis that pheromones are ancestors of present-day hormones and that they arose very early during the evolutionary history as local cellular signals in unicellular organisms (primordial exocrine system).

Essentially, these bioregulators act as autocrine and paracrine factors (Vallesi *et al.* 1995). For example, cAMP regulates the metabolism of prokaryotes (autocrine action) (Mackman & Sutherland 1965), and regulates many activities in unicellular eukaryotes (paracrine action) (Schapp 1984). Moreover, cAMP plays a dual role as intra- and intercellular messenger in *Dictyostelium discoideum* (Gerish 1987).

What are the origins of this dual role of cAMP?

If a bioregulator acts as inter- and intracellular messenger receptor transformations must be involved. For instance, the action of cAMP as second messenger suggests that an internalization process takes place, because in *Escherichia coli* cAMP is produced in the same cell in which it exerts its effects (autocrine action) and forms a complex with an extracellular specific protein, the cAMP-receptor protein (CRP) (Mackman & Sutherland 1965). This protein is homologous to protein kinases, which are intracellular cAMP receptors in unicellular eukaryotes (Weber *et al.* 1982, Saxe *et al.* 1991).

It is obvious that with the development of multicellularity, cAMP cannot act as a cell-to-cell messenger because of its intrinsic chemical instability, and can only carry information as an intracellular second messenger by means of cytosolic cAMP-dependent protein kinases (Weber *et al.* 1982).

The presence of membrane and cytosolic cAMP receptors suggests the development of transitional stages during evolution. Clearly, each form of cAMP/cAMP receptor unit is linked to a determined facet within the evolutionary triptych of bioregulator-receptor interaction including: (i) autocrine (membrane form of cAMP receptor, cAMP=first messenger); (ii) paracrine (membrane and cytosolic forms of cAMP receptor, cAMP=first and second messenger); (iii) endocrine (cytosolic form of cAMP receptor, cAMP=second messenger).

The transitional taxonomic level on the cAMP receptor evolution may be represented today by *Dictyostelium discoideum*, an intermediate stage displaying: (i) a level of organization that is

TABLE 2. Ubiquity of information molecules with structural homology and/or functional analogy to those of vertebrates

Organism	Binding protein for:	K_d	Bioregulator	G-protein	Reference
Archaeobacteria					
<i>Halobacterium halobium</i>	—	—	—	59 kDa	Schimz <i>et al.</i> (1989)
Eubacteria					
<i>Escherichia coli</i>	—	—	—	35 kDa	Ahnn <i>et al.</i> (1986)
—	—	—	Insulin	—	Le Roith <i>et al.</i> (1981a)
—	—	—	Somatostatin	—	Le Roith <i>et al.</i> (1985a)
<i>Clostridium perfringens</i>	—	—	Thyrotrophin	—	Macchia <i>et al.</i> (1967)
<i>Progenitor cryptocides</i>	—	—	Human choriogonadotrophin	—	Maruo <i>et al.</i> (1979)
<i>Yersinia enterocolitica</i>	Thyrotrophin	4.2×10^{-8} M	—	—	Weiss <i>et al.</i> (1983)
<i>Pseudomonas testosteroni</i>	Testosterone	6.7×10^{-9} M	—	—	Watanabe <i>et al.</i> (1973a)
<i>Pseudomonas maltophilia</i>	Human choriogonadotrophin	2.3×10^{-9} M	—	—	Richert & Ryan (1977)
Fungi					
<i>Dictyostelium discoideum</i>	—	—	—	42 kDa	Leichtling <i>et al.</i> (1981)
<i>Saccharomyces cerevisiae</i>	—	—	—	54 kDa	Brock <i>et al.</i> (1985)
—	17 β -Oestradiol	3.0×10^{-9} M	17 β -Oestradiol	—	Feldman <i>et al.</i> (1982, 1984)
—	GnRH	1.0×10^{-6} M	Alpha factor homologous to GnRH	—	Loumaye <i>et al.</i> (1982)
<i>Neurospora crassa</i>	—	—	Insulin	—	Le Roith <i>et al.</i> (1980)
<i>Trichophyton mentagrophytes</i>	Progesterone	6.8×10^{-8} M	—	—	Schar <i>et al.</i> (1986)
<i>Paracoccidioides brasiliensis</i>	17 β -Oestradiol	1.5×10^{-8} M	—	—	Loose <i>et al.</i> (1983)
<i>Candida albicans</i>	Corticosterone	7.2×10^{-9} M	—	—	Loose & Feldman (1982)
Protozoa					
<i>Trypanosoma cruzi</i>	—	—	—	45 kDa	Eisenschlos <i>et al.</i> (1986)
<i>Tetrahymena pyriformis</i>	—	—	Relaxin	—	Schwabe <i>et al.</i> (1983)
—	—	—	Insulin	—	Le Roith <i>et al.</i> (1980)
—	—	—	Adrenocorticotrophic hormone, B-endorphin, somatostatin	—	Le Roith <i>et al.</i> (1983)
—	—	—	Adrenaline, noradrenaline	—	Janakidevi <i>et al.</i> (1966)
Platyhelminthes					
Planarians	—	—	Dopamine, noradrenaline	—	Welsh & King (1970)
Annelids					
<i>Glycera convoluta</i>	—	—	Adrenaline, noradrenaline, dopamine, serotonin	—	Manaranche & L'Hermite (1973)
Mollusca					
<i>Anodonta cingnea</i>	—	—	Insulin	—	Plisetskaya <i>et al.</i> (1978)
<i>Mytilus edulis</i>	—	—	Testosterone, 17 β -oestradiol	—	De Longcamp <i>et al.</i> (1974)
<i>Lymnaea stagnalis</i>	—	—	Somatostatin	—	Grimm-Jorgensen (1983)

TABLE 2. Continued

	Binding protein for:	K_d	Bioregulator	G-protein	Reference
Echinoderms					
<i>Pisaster ochraceus</i>	—	—	17 β -Oestradiol	—	Botticelli <i>et al.</i> (1960)
Arthropods					
<i>Dytiscus marginalis</i>	—	—	Oestrogens, androgens	—	Schildknecht <i>et al.</i> (1967)
<i>Calliphora vomitoria</i>	—	—	11-deoxycorticosterone	—	Schildknecht <i>et al.</i> (1966)
<i>Drosophila melanogaster</i>	—	—	Insulin	—	Duve <i>et al.</i> (1979)
			—	40 kDa	Thambi <i>et al.</i> (1989)
Urochordates					
<i>Ciona intestinalis</i>	—	—	Somatostatin	—	Falkmer <i>et al.</i> (1978)
Plants					
<i>Pinus silvestris</i>	—	—	Testosterone	—	Saden-Krekula <i>et al.</i> (1971)
Spinach, tobacco	—	—	Somatostatin	—	Le Roith <i>et al.</i> (1985b)
Wheat	—	—	Opioids	—	Zioudrov <i>et al.</i> (1979)

GnRH, gonadotrophin releasing hormone.

transitional between prokaryotic and multicellular organisms; (ii) *D. discoideum* has both membrane and cytosolic forms of cAMP receptors; (iii) in this species cAMP acts as a paracrine bioregulator (first messenger) and intracellular regulatory molecule (second messenger) (Table 4).

It seems likely that in the development of transitional stages such as *D. discoideum*, the earliest membrane receptors were the predecessors of cytosolic receptors. There are other examples of transitional stages; for example, progesterone, testosterone and gonadotrophins can act through both membrane and cytosolic receptors (Baulieu *et al.* 1978, Rao & Chegini 1983, Diez *et al.* 1984).

Briefly, contemporary endocrine bioregulators (hormones, neuroactive materials) were probably, in the past, exocrine information molecules (food signals, pheromones), and their contemporary clearly defined endocrine functions (sexual reproduction, developmental processes) are more a product of target tissue specialization (e.g. transduction mechanisms) than a consequence of changes in the structure of the bioregulators themselves.

These views are supported by several observations: (i) the most effective signals for the chemotactic response in *E. coli* (maltose, glucose, serine, aspartate) are used as nutritive molecules (Adler 1969) in contrast to more specialized bioregulators (neuroactive compounds, hormones); (ii) the receptors or binding proteins in unicellular organisms display clear threshold values for chemotaxis towards a particular bioregulator (Adler 1975), which resemble the dissociation constant (K_d) of vertebrate-type bioregulators (Table 2). For example the threshold value of reduced glutathione (GSH), a feeding response activator (Lenhoff 1968, Colasanti *et al.* 1995), in *Hydra littoralis*, a member of one of the most ancient phyla among multicellular organisms, the Coelenterates, is 10^{-9} M.

GSH has, phylogenetically, the characteristics of a transitional stage among food signals and contemporary bioregulators (hormones, pheromones) because: (i) it plays a dual role acting in *Hydra* both as a chemical signal for detection of food sources, and as a neuromodulator of behavioural feeding response; (ii) like contemporary bioregulators, GSH is not used as a nutritive molecule.

In addition, primitive peptides and ancestral lipid molecules (sterols, steroids) were probably all growth promoting factors as a consequence of their intrinsic nutritional value. For example, *Pseudomonas testosteroni* is a prokaryote capable of utilizing steroids as their only carbon source (Watanabe *et al.* 1973a,b).

It is thus important to note a plausible relationship among some enzymes involved in the

TABLE 3. Heterotrophic effects of some bioregulators

Bioregulator	Organism	Effects	Reference
17β-Oestradiol	Vertebrates	Maturation of accessory sexual tissues	Geuns (1978)
	Plants	Induction of flowering, sex determination	Geuns (1978)
	<i>Opalina ranarum</i> (Protozoan)	Cause encystment	El Mofly & Smyth (1964)
	<i>Trypanosoma cruzi</i> (Protozoan)	Growth inhibition	A M Stoka (unpublished observation)
	<i>Coccidioides immitis</i> (Fungi)	Growth stimulation	Druz <i>et al.</i> (1981)
Progesterone	<i>Paracoccidioides brasiliensis</i> (Fungi)	Growth inhibition	Loose <i>et al.</i> (1983)
	<i>Trichophyton mentagrophytes</i> (Fungi)	Growth inhibition	Schar <i>et al.</i> (1986)
	<i>Tetrahymena pyriformis</i> (Protozoan)	Growth inhibition	Csaba & Fulop (1987)
	<i>Xenopus laevis</i> (Amphibia)	Induction of meiotic division	Baulieu <i>et al.</i> (1978)
		Regulation of: (a) oxidative metabolism, (b) growth	Tata (1964)
Thyroxine	Mammals	Regulation of metamorphosis	Tata (1964)
	Amphibians	Growth stimulation	Csaba & Nemeth (1980)
	<i>Tetrahymena pyriformis</i> (Protozoan)		
Prolactin	Mammals	Milk secretion	Bern & Nicoll (1969)
	Birds	Crop-milk production	Bern & Nicoll (1969)
Adipokinetic hormone	Insects	Lipid mobilization	Stone <i>et al.</i> (1976)
	Crustaceans	Pigment concentration	Stone <i>et al.</i> (1976)
Juvenile hormone	Insects	Regulation of: (a) reproduction, (b) metamorphosis	Stoka (1987)
	<i>Trypanosoma cruzi</i> (Protozoan)	Growth inhibition	Stoka <i>et al.</i> (1990); Stoka (1996)
	Mammals	Inhibition of testicular steroidogenesis	Vladusic <i>et al.</i> (1994)
20-OH-ecdysone	Insects	Moulting hormone	Stoka (1987)
	<i>Trichonympha sp</i> (Protozoan)	Growth stimulation and sexual differentiation	Cleveland (1959)

TABLE 4. Evolutionary transition of cAMP-receptors: from membrane to cytosolic environment

Organization level	Organism	Type of c-AMP receptor	c-AMP function	Interaction c-AMP/c-AMP receptor
*Prokaryote	<i>Escherichia coli</i>	Membrane form	First messenger	Autocrine
†Eukaryote unicellular	<i>Dictyostelium discoideum</i>	Membrane and cytosolic forms	First and second messenger	Paracrine
‡Eukaryote multicellular	Vertebrates and invertebrates	Cytosolic form	Second messenger	Endocrine

*Mackman & Sutherland (1965); †Gerish (1987); Saxe *et al.* (1991); ‡Weber *et al.* (1982).

metabolism of bioregulators, and the origin of receptors. An interesting point of view is that binding and catalytic activities of some cell surface receptors arose from primitive enzymes. This hypothesis is supported by the presence of some transitional stages among enzymes and receptors: (i) the transport of testosterone in membrane vesicles of *P. testosteroni* is coupled to membrane bound 3β - and 17β -hydroxysteroid dehydrogenases (Watanabe & Po 1976); (ii) a membrane form of guanylate cyclase in mammals acts as a receptor for atrial natriureic peptide (Chinkers *et al.* 1989); and (iii) the cytosolic receptor for salicylic acid in plants has catalase activity (Chen *et al.* 1993).

Furthermore, some point mutations in the binding site of an enzyme enhanced its substrate binding capacity although they produced a catalytically non-productive species (Craik *et al.* 1985).

The catalytic and binding capacities of prokaryotes were probably employed in a different fashion to their contemporary hormonal ones in multicellular organisms and represented a vestige of Precambrian times (exaptation process). Some advantageous changes in the structure of prokaryotic enzymes involved in the metabolism of nutritive substrates (e.g. 3β - and 17β -hydroxysteroid dehydrogenases of *P. testosteroni*) may have 'filtered' through the natural selection process, and evolved into other functions such as autocrine, paracrine and/or endocrine signal receptors to facilitate regulation of intracellular processes. In this evolutionary scenario a different type of regulation may be considered. A bioregulator can be produced and act within the same cell (intracrine regulation) (O'Malley 1989). It is known that certain unicellular organisms biosynthesize steroid hormones and contain intracellular steroid receptors. Examples include *Saccharomyces cerevisiae* and *Candida albicans* (Loose *et al.* 1981, Loose & Feldman 1982, Feldman *et al.* 1982, 1984).

If the first unicellular organisms possessed few multifunctional molecules, probably nutritional compounds would serve both as metabolic and

regulatory elements. It is possible then, that intracrine regulation could be a transitional stage between regulation through metabolic substrates (nutritive line) and hormonal modulation (regulatory line).

Primitive members of the contemporary steroid receptors family were possibly membrane enzymes which bound environmental nutritional molecules (steroids, sterols), and their prime regulatory function was to control intracellular metabolism.

Such relationships between food signals and contemporary bioregulators do not preclude other parallel original functions of bioregulators in defence functions.

The primary role of ancestral bioregulators in unicellular organisms was a nutritional one, but the adaptive radiation among molecules produced compounds that were inhibitors (e.g. toxins) of enzymes involved in the growth of competitive species.

PHYLOGENETIC ASPECTS OF BIOREGULATOR-RECEPTOR RELATIONSHIP

Some ligand-binder relationships (substrate-enzyme, antigen-antibody, bioregulator-receptor) must be restricted to an interaction with specific and small regional domains within their structures, as exemplified by insulin and its receptor (Pullen *et al.* 1976).

The chemical structure of bioregulators has been well conserved through evolution and there are high degrees of homology within the system (Table 2). In addition, the divergent functions of bioregulators with a similar structure (e.g. progesterone/cortisol) pose a number of questions including the following.

Have receptors evolved in a similar way? How similar and how different are receptors for different bioregulators?

A case in point is that the amino acid sequences of nuclear thyroid hormone receptors display homology with a similar domain of several steroid

hormone receptors (Weinberger *et al.* 1986). Thus, the relationship between steroid hormone receptors and thyroid hormone receptors suggests that all these receptor structures, including the v-erb-A oncogene product of the avian erythroblastosis virus (AEV), may be a superfamily of information molecules that have evolved in several divergent lines during their phylogeny. However, the existing homologies between the thyroid hormone receptors and steroid hormone receptors, vitamin D₃, aldosterone, cortisol, testosterone, progesterone or oestradiol did not seemingly compete (Weinberger *et al.* 1986). Obviously, the secondary, tertiary and quaternary structures of the thyroid hormone receptor increase specificity for thyroid hormone.

What is the origin of the bioregulator–receptor specificity?

The origin of cell specificity for a determined bioregulator depends on the critical period of receptor maturation which determines its binding properties (Blazquez *et al.* 1976) and which may be influenced or altered by the action of a similar chemical structure (Csaba 1980).

On the other hand, the superfamily of steroid receptors, besides receptors binding thyroid hormones, retinoic acid or steroid hormones, contains orphan receptors for which no ligand is known (O'Malley 1989, Escriva *et al.* 1997). It seems likely that the number of ancestral steroid receptors (enzymes?) was expanded by the presence of immature receptors which adopted different binding site configurations with high specificity for particular ligands, as occurs with the evolution of antibody catalysis (Wedemayer *et al.* 1997).

Another aspect involved in the origin of bioregulator–receptor interaction is the structural similarity between secreted bioregulators and chemical components of cells. It has been suggested that pheromones and hormones evolved from membrane components (Kochert 1978, Pfeffer & Ullrich 1985). Accordingly, there are two hypotheses to explain the origins of bioregulator–receptor units.

Hypothesis 1: polypeptide hormones are produced by proteolytic cleavage of membrane proteins of secondary lysosomes (Hales 1985)

The peptide hormone, epidermal growth factor (EGF), is cleaved from membrane protein (Kaback 1985). In addition, bioregulators and their receptors may evolve as a result of changes in the DNA by gene duplication and subsequent point mutational events. Some genes encoding receptor–bioregulator units began as single genes encoding single

polypeptide molecules. Gene splitting and different gene expressions may have induced the synthesis of bioregulators and their receptors in different cell types (Niall 1982, Kaback 1985).

Hypothesis 2: the presence of a bioregulator is an essential condition for receptor formation (the theory of signal-imprinting, Csaba 1980)

During this process, there is likely to be initial contact between a chemical signal and a potential receptor region of the cell membrane, and induction of shape complementarity and hydrophobicity of the interacting surfaces may take place (Tainer *et al.* 1984).

However, it is not only the bioregulator–receptor unit which determines the quality of physiological responses under its control. Phylogenetically transducing systems involve trimeric G-proteins, identified in various prokaryotes (Ahnn *et al.* 1986, Schimz *et al.* 1989) and eukaryotes (Namba *et al.* 1983, Firtel *et al.* 1989, Whiteway *et al.* 1989).

In addition to the homologies among steroid receptors and thyroid hormone receptors (Weinberger *et al.* 1986), there is another type of homology – that of G-proteins. Key observations are: (i) the prostaglandin E receptor subtype EP3 and the thromboxane A₂ receptor belong to the large superfamily of receptors coupled to G-proteins which consist of seven transmembrane domains (Namba *et al.* 1983) and are isoforms of rhodopsin (Franke *et al.* 1990) and dopamine receptors (Monsma *et al.* 1989); (ii) the sequence and size variation among receptor domains whereby receptors bind G-proteins (Monsma *et al.* 1989) suggest widespread diversity within the G-protein family. Thus, different physiological responses of receptor isoforms are structurally based (Fig. 3).

In addition, there is other evidence for conservation and functional versatility of information molecules: (i) the pheromone signalling pathway in *S. cerevisiae* is activated by an heterologous transducing system including the human β -adrenergic receptor and the rat Gs-protein (King *et al.* 1990); (ii) *S. cerevisiae* has a primitive steroid receptor system analogous to the mammalian oestradiol receptor unit (Feldman *et al.* 1982, 1984); (iii) a bioregulator can interact with autocrine and paracrine receptors to accrue more physiological functions (Mackman & Sutherland 1965, Gerish 1987, Saxe *et al.* 1991, Vallesi *et al.* 1995); (iv) a receptor in combination with homologous bioregulators can regulate the same physiological response in a determined target tissue (Stone *et al.* 1976, Mordue & Stone 1976, 1977); (v) a receptor can bind different bioregulators and elicit varying patterns of gene activation but with the same effects

FIGURE 3. Different receptor isoforms can produce more than one physiological response. B, Bioregulator; R, membrane receptor; T, transducer (e.g. G-protein); E, effector (e.g. cyclases, phospholipases); SM, second messengers (e.g. cAMP); SMC, second messenger cascades (e.g. inositol triphosphate cascade); PR, physiological response; CM, cell membrane; EE, extracellular environment; IE, intracellular environment. The G-protein diversity probably results from multiple genes and/or alternative RNA splicing generating multiple isoforms from a single gene. Possible scenarios: (a and b) receptors are coupled to different transducers; (a and c) different domains of a determined receptor interact with the same transducer but with an analogous effector. This effector may react with the same or with a different second messenger; (a and d) receptor isoforms activate different effectors.

in a particular tissue (Yang *et al.* 1996, Paech *et al.* 1997).

SOME HYPOTHESES TO EXPLAIN THE EVOLUTION OF INFORMATION MOLECULES

A basic question is: were contemporary bioregulators (hormones, neuroactive compounds, growth factors, pheromones) primary chemical messengers such as food signals and defensive compounds?

Sterols have probably played an important function as growth bioregulators in the Precambrian period, 1000–3000 $\times 10^6$ years ago. Some evidence supports this hypothesis: (i) cellular life may have been present about 3000 $\times 10^6$ years ago as fermenting bacteria (Schwemmler 1984); (ii) sterol biosynthesis existed at a very early stage in some organisms such as the Cyanobacteria (blue-green algae, *Nostoc*, *Spirulina*) (Nes & McKean 1977); (iii) some sterols participate in the activation of oocyte meiosis (Byskov *et al.* 1995) – a possible feature of ancestral bioregulators was the control of cell division; (iv) the presence of both binding and

catalytic activities for vertebrate-type steroid hormones in aerobic bacteria (Watanabe *et al.* 1973a,b, Watanabe & Po 1976).

Indeed there are important differences in structural conservation during the phylogeny of information molecules. Some bioregulators such as steroid and thyroid hormones are rigorously conserved because they result from enzymic cascades and require almost absolute substrate specificity. In contrast others (somatomammotrophins, pituitary glycoproteins, secretins) are direct gene products and will tolerate change providing the 'active' (receptor activating) portion of the peptide is not changed and variation can be tolerated (Henderson 1997).

In some cases, the structural evolution of information molecules produced a functional versatility of a hormone. For example, prolactin has acquired different functions among vertebrates: (i) osmoregulatory activity in fish; (ii) growth-promoting activity in tetrapods; (iii) metamorphic actions in amphibians, and (iv) lactogenic activity for neonatal nutrition in mammals and birds (Bern & Nicoll 1969).

Correlated with these physiological acquisitions, comparative studies on structural and functional bioregulator–receptor interactions reveal that both prolactin and prolactin receptors have undergone considerable changes (White & Nicoll 1979), although there is no strict relationship between functional versatility and the number of chance substitutions in the structure of informational molecules.

A most striking example in this area is the structural change found in naturally occurring insulins among vertebrates. These insulins have different affinities for the insulin receptor in mammalian tissues (Muggeo *et al.* 1979) and they differ 50- to 100-fold in their potency (Blundell *et al.* 1972).

At the level of specificities among ligands and binders, some bioregulator–receptor units seem to be in a transitional evolutionary stage according to their cross reactions, as occurs with arthropod neurohormones (Fig. 4, Table 5). In contrast, other bioregulator–receptor units such as the glucagon/glucagon receptor unit (Blundell & Humbel 1980) display maximum specificity. It is thus possible that different bioregulator–receptor units in a particular species evolved at different rates and times, as occurred with some morphological characteristics, a process termed mosaic evolution (De Beer 1954); there are several examples for different molecules (Wilson *et al.* 1977). From a functional point of view, the prolactin/prolactin receptor unit has evolved more rapidly than the insulin/insulin receptor unit

FIGURE 4. Structural homologies among arthropod neurohormones. The primary structures of red pigment concentrating hormone of prawn (RPCH, Crustacea, octapeptide), adipokinetic hormone of locust (AKH, Insecta, decapeptide) and periplanetin of cockroach (CC-1, Insecta, octapeptide) suggest that they evolved from a common precursor by gene duplication and subsequent point mutational events (*); then they diverged functionally. *AKH: Serine (SER) — Threonine (THR) (amino acid position 5); CC-1: Leucine (LEU) — Valine (VAL) (amino acid position 2); RPCH: Asparagine(ASN) — Glycine (GLY) (amino acid position 7).

as judged by the number of known physiological functions acquired during vertebrate evolution.

In addition, the taxonomic distribution of insulin and prolactin differs (Fig. 5) and does not correlate with their functional versatility. In this regard, it may be that chemical communication arose many times during phylogeny and primordial information molecules, with functional and/or structural characteristics different to present-day bioregulator–receptor units, may have appeared and disappeared as a result of changes in the expression of genetic information (point mutations, gene duplication, alternative splicing, transpositions, etc.). The very nature of the regulation of genetic transcription would indicate that the genome of contemporary biota must contain DNA regions with information for potential unexpressed bioregulator–receptor units, and therefore the specific group of present-day information molecules may be only one of several lineages.

TABLE 5. Effects of arthropods neurohormones

Neurohormone	Pigment concentrating activity in erythrophores (Crustacea)	Hyperglycaemic activity in fat body cells (Insecta)	Adipokinetic activity in fat body cells (Insecta)
†Adipokinetic hormone (AKH)	Yes*	Yes*	Yes
‡Periplanetin (CC-1)	—	Yes	Yes*
§Red pigment concentrating hormone	Yes	—	Yes*

Cross-reactions (*) were observed among arthropod neurohormones. Probably binding and transduction activities of neurohormone receptors could have evolved independently. The evolution of arthropod neurohormones and their receptors seems to be an example in which specificity has not reached a maximum (transitional or immature stage).

†Mordue & Stone (1976, 1977); Stone *et al.* (1976); ‡Hanaoka & Takahashi (1976); §Mordue & Stone (1976, 1977); Ferlund (1974).

Indeed, these non-translated DNA regions (introns) are, in terms of genetic information, the remnant of phylogenetic history and the building blocks of current and future evolution.

This hypothesis is supported by ectopic endocrine activity of patients with neoplasia. It was observed that the occasional disruption of normal cell differentiation can produce nonendocrine tumours, capable of synthesizing a great amount of hormones (e.g. pheochromocytoma) (Baylin & Mendelsohn 1980). Such complicated and fascinating processes reflect a regression towards a primordial cellular stage; under such conditions it seems that there is a return to a cellular pluripotentiality for the biosynthesis of information molecules. Such potential information molecules may arise from ‘hidden’ sequences of DNA regions (introns) and developed unknown (extinct or new) bioregulator signalling mechanisms. Evolution in a sense can go into reverse to re-express earlier chemical communication systems.

It is also plausible that present-day chemical communication systems may be superior to primordial systems, but they may not necessarily always be so under some conditions.

CONCLUSIONS

The inherent similarity of mechanisms involved in bioregulator–receptor interactions reveals that glandular and nervous systems (endocrine systems) may have a common origin from a pheromonal system (primordial exocrine system) of unicellular organisms.

This hypothesis can explain many phenomena: (i) vertebrate-type hormones acting in unicellular organisms as pheromones that cause regulatory effects (Tables 2 and 3); (ii) the presence of vertebrate-type bioregulators (gastrin, somatostatin, prolactin) in classical exocrine fluids such as saliva, gastric juice and other humoral secretions (Le Roith

FIGURE 5. Conservation of some bioregulators during biological evolution. Some properties of bioregulator-receptor units suggest that they may have evolved by selection of some other function, and then diverged functionally. Certainly, the conserved specific domains of bioregulator-receptor units and their presence in different taxonomic groups are consistent with a natural selection event to ensure survival and reproduction of new life forms (adaptation), but in certain cases (Riedel *et al.* 1986) their final physiological function is largely unknown (exaptation). ■ Present-day distribution of different bioregulators through different taxonomic levels. 1, Insulin; 2, glucagon; 3, prolactin; 4, noradrenaline; 5, potential but not expressed bioregulators; 6, juvenile hormone and juvenile hormone analogues. Juvenile hormone activity has been found in protozoans (Fisher & Sandborn 1962), arthropods (Judy *et al.* 1973), vertebrate tissues (Gilbert & Schneiderman 1958, Williams *et al.* 1959) and plants (Slama & Williams 1965, Bowers & Nishida 1980)); 7, oestradiol; 8, 20-hydroxyecdysone; 9, somatostatin. □ Extinct or unexpressed pathways of bioregulator biosynthesis in contemporary species. P, Prokaryotes; E, eukaryotes; U, unicellular; M, multicellular; H, heterotrophs; A, autotrophs; I, invertebrates; V, vertebrates.

et al. 1986); (iii) the ubiquity of insulin in extrapancreatic tissues of mammals (brain, liver, cultured lymphocytes and fibroblasts) (Rosenzweig *et al.* 1980), insects (Duve *et al.* 1979), annelids (Le Roith *et al.* 1981b) and unicellular organisms (Le

Roith *et al.* 1980, 1981b); (iv) the overlap of brain and gut bioregulators in mammals (Zimmerman 1979) and insects (Duve & Thorpe 1981); (v) the biosynthesis of gastrointestinal hormones in neural, endocrine and paracrine cells (Grosman 1979); (vi) gonadal steroid hormones acting as defensive agents (Schildknecht *et al.* 1966, 1967, Loose *et al.* 1983, Schar *et al.* 1986); (vii) the presence of specific binding of insulin to the unicellular alga *Acetabularia mediterranea* (Legros *et al.* 1975); (viii) the ubiquitous distribution of thyrotrophin releasing hormone in the animal kingdom (Henderson 1997) and plants (Morley *et al.* 1980, Jackson 1981).

It is suggested that endocrine secretory mechanisms (glandular, nervous) have a common origin and are products of phylogenetic and evolutionary processes from unicellular organisms (primordial exocrine bioregulators) to multicellular organisms (contemporary exocrine and endocrine bioregulators).

On the other hand, receptor binding activities seem to have arisen as a result of modifications in the structure of enzymes. Within this hypothesis a divergent evolution is possible since enzyme binding activity need not always co-evolve with its intrinsic catalytic activity. Such an evolutionary mechanism enables receptors to be created without the development of additional binding sites (Stone *et al.* 1976, Shemshedini & Wilson 1990, Lee *et al.* 1992).

It is not surprising that receptor evolution, as with other evolutionary characteristics, required millions of years during which some transitional stages (e.g. hormonal receptors with catalytic activity) predominated. Enzymes are thus the key to understanding the origin and evolution of both enzymes and receptor binding activities. Some basic questions are obviously raised: how similar are the binding regions of receptors and enzymes for a particular bioregulator? Is the origin of receptor and enzyme binding activities, for a particular bioregulator, a product of convergent or divergent evolutionary processes?

Although evidence is scanty, it is possible that by mutational and natural selection processes a bioregulator-receptor unit arose through initial divergent evolution followed by a convergent process. Convergent and divergent evolution have been noted in some eukaryotic phosphorylases (Hwang & Fletterick 1986). Figure 6 illustrates possible sequences within the evolutionary history of bioregulator-receptor units.

The evolutionary development of chemical communication systems reflects parallel evolution of all information molecules (bioregulators, receptors,

FIGURE 6. Hypothetical sequence of evolutionary transitions from a potential signal-receiver unit to bioregulator-receptor interaction. Evolutionary stages: 1, potential signal-receiver unit; 2, substrate-enzyme interaction; 3, pheromone-membrane receptor interaction; 4, neurotransmitter or hormone-receptor interaction; 5, bioregulator (pheromone, hormone, neurotransmitter)-nuclear receptor interaction. Initially, in this schematic representation, a potential signal-receiver unit (stage 1) may have evolved into two divergent models: (i) enzyme (membrane form)-substrate (nutritive line) (stage 2); (ii) membrane receptor-pheromone (regulatory line) (stage 3). After this divergent process, a convergent evolution is possible; for instance the receptor binding activities arose from enzymes involved in the metabolism of nutritive substrates as a result of: (i) point mutations in the binding site (transition c substrate specificity enhancement and catalytic activity diminution) (Craik *et al.* 1985); (ii) altered binding specificity may also influence interactions of some molecules (nutritive substrates, transition molecules) with membrane receptors which acquire a pheromonal status. On the other hand, these transition molecules may have evolved into toxins which may serve as enzyme inhibitors or enzyme activators. Transitions: a, origin of signal-receiver units through genetic or imprinting mechanisms; b, origin of endocrine bioregulators (neurotransmitters, hormones, etc.) through the development of multicellularity; c, hormonal and pheromonal receptors arose from primitive enzymes; d, probable relationship between nutritive substrates and food signals; e, transition molecules (pheromone-like compounds) arose from advantageous changes in the structure of food signals; f, receptor internalization process: from membrane receptors to soluble intracellular receptors. BBB, biological building blocks; CM, cell membrane; DNA-ReEl, DNA-response element, R, receptor; H, hormone; P, pheromone; NS, nutritive substrate; E, enzyme; B, bioregulator (hormone, pheromone, etc.), TM, transition molecule; FS, food signal; T, toxin; N, neurotransmitter.

transducers, enzymes, second messengers, second messenger cascades, etc.), which are sometimes co-ordinated and essential processes (adaptation) and are sometimes opportunistic and random events (exaptation).

Despite extensive accumulation of information over the last 20 years, many important gaps remain in our knowledge of phylogenetic relationships and genetic processes therein.

Virtually nothing is known about: (i) the effects of mutations in different kinds of genes (multiple genes, simple non-overlapping genes) that can be quite different according to their primary function, (ii) whether it is possible to describe the exact physico-chemical conditions and frequencies of new adaptations and exaptations.

Molecular endocrinology is now at a stage when some of these and other questions can be addressed. The author expects that this review will stimulate new routes of thinking in research teams that work in the area.

ACKNOWLEDGEMENTS

This work was carried out with financial assistance from Consejo Nacional de Investigaciones Cientificas y Tecnicas (CONICET), Argentina.

I thank Drs Luis Mayorga and Armando Parodi for useful comments on the manuscript. I would also like to express my thanks to Prof. I Henderson for his assistance in the revision of the manuscript.

REFERENCES

- Adler JA 1969 Chemoreceptors in bacteria. *Science* **166** 1588–1597.
- Adler JA 1975 Chemotaxis in bacteria. *Annual Review of Biochemistry* **44** 341–356.
- Ahn J, March PE, Takiff HE & Inouye M 1986 A GTP-binding protein of *Escherichia coli* has homology to yeast RAS proteins. *Proceedings of the National Academy of Sciences of the USA* **83** 8849–8853.
- Barksdale AW 1967 The sexual hormones of the fungus *Achlya*. *Annals of the New York Academy of Sciences* **144** 313–319.
- Baulieu EE, Godeau F, Schorderet M & Schorderet-Slatkine S 1978 Steroid-induced meiotic division in *Xenopus laevis* oocytes: surface and calcium. *Nature* **275** 593–598.
- Baylin SB & Mendelsohn G 1980 Ectopic (inappropriate) hormone production by tumors: mechanisms involved and the biological and clinical implications. *Endocrine Reviews* **1** 45–77.
- Bentley PJ 1976 *Comparative Vertebrate Endocrinology*. New York: Cambridge University Press.
- Bern HA & Nicoll CS 1969 The taxonomic specificity of prolactins. *Progress in Endocrinology Excerpta Medica, Amsterdam* 433–439.
- Blazquez E, Rubalcava B, Montesano R, Orci L & Unger RH 1976 Development of insulin and glucagon binding and the adenylate cyclase response in liver membranes of the prenatal, postnatal and adult rat: evidence of glucagon 'resistance'. *Endocrinology* **98** 1014–1023.
- Blundell TL & Humbel RE 1980 Hormone families: pancreatic hormones and homologous growth factors. *Nature* **287** 781–787.
- Blundell TL, Dodson GG, Hodgkin DC & Mercol DA 1972 Insulin: the structure in the crystals and its reflection in chemistry and biology. *Advances in Protein Chemistry* **26** 279–402.
- Bonner JT 1971 Aggregation and differentiation in the cellular slime molds. *Annual Review of Microbiology* **25** 75–92.
- Botticelli CR, Hisaw FL & Wotiz HH 1960 Estradiol-17 β and progesterone in ovaries of starfish (*Pisaster ochraceus*). *Proceedings of the Society of Experimental Biology and Medicine* **103** 875–877.
- Bowers WS & Nishida R 1980 Juvocimenes: potent juvenile hormone mimics from sweet basil. *Science* **209** 1030–1032.
- Brock D, Samly N, Fasano O, Fujiyama A, Tamanos F, Northup J & Wigler M 1985 Differential activation of yeast adenylate cyclase by wild-type and mutant RAS proteins. *Cell* **41** 763–769.
- Buetow DE & Levedahl BH 1964 Responses of microorganisms to sterols and steroids. *Annual Review of Microbiology* **18** 167–194.
- Butenandt A 1963 Bombycol, the sex attractive substance of the silkworm *Bombyx mori*. *Journal of Endocrinology* **27** IX–XVI.
- Butler CG 1967 Insect pheromones. *Biological Reviews* **42** 42–87.
- Byskov C, Andersen CY, Nordholm L, Thøgersen H, Guoliang X, Wassmann O, Andersen JB, Guddal R & Roed T 1995 Chemical structure of sterols that activate oocyte meiosis. *Nature* **374** 559–562.
- Chang C, Kwok SF, Bleecker AB & Meyerowitz EM 1993 Arabidopsis ethylene-response gene ETR1: similarity of product to two-components regulators. *Science* **262** 539–544.
- Chen Z, Silva H & Kleissig DF 1993 Active oxygen species in the induction of plant systemic acquired resistance by salicylic acid. *Science* **262** 1883–1886.
- Chinkers M, Garbers DL, Chang MS, Lowe DG, Chin H, Goeddel DV & Schulz S 1989 A membrane form of guanylate cyclase is an atrial natriuretic peptide receptor. *Nature* **338** 78–83.
- Cleveland LR 1959 Sex induced with ecdysone. *Proceedings of the National Academy of Sciences of the USA* **64** 747–753.
- Colasanti M, Lauro GM & Venturini G 1995 NO in *Hydra*. *Nature* **374** 505.
- Craik CS, Largman C, Fletcher T, Rocznik S, Barr PJ, Fletterick R & Rutter WJ 1985 Redesigning trypsin: alteration of substrate specificity. *Science* **228** 291–297.
- Crisp DJ & Meadows PS 1962 The chemical basis of gregariousness in cirripides. *Proceedings of the Royal Society (B)* **156** 500–520.
- Csaba G 1980 Phylogeny and ontogeny of hormone receptors: the selection theory of receptor formation and hormonal imprinting. *Biological Reviews* **55** 47–63.
- Csaba G & Nemeth G 1980 Effect of hormones and their precursors on Protozoa. The selective responsiveness of *Tetrahymena*. *Comparative Biochemistry and Physiology* **65B** 387–390.
- Csaba G & Fulop K 1987 Steroid hormone (prednisolone) influence on the unicellular *Tetrahymena* (an electron microscope study). *Acta Protozoologica* **26** 233–236.
- De Beer G 1954 *Archaeopteryx* and evolution. *Adv. Science* **11** 160–170.

- De Longcamp D, Lubet P & Dubrowsky M 1974 The 'in vitro' biosynthesis of steroids by the gonads of the mussel (*Mytilus edulis*). *General and Comparative Endocrinology* **22** 116–127.
- Diez A, Sancho MJ, Egana M, Trueba M, Marino A & Maccarella JM 1984 An interaction of testosterone with cell membranes. *Hormone and Metabolic Research* **16** 475–477.
- Donta ST & Moon HW 1974 Detection of heat-labile *Escherichia coli* enterotoxin with the use of adrenal cells in tissue culture. *Science* **183** 334–336.
- Drutz DJ, Huppert M, Sun SH & McGuire WL 1981 Human sex hormones stimulate the growth and maturation of *Coccidioides immitis*. *Infection and Immunity* **32** 897–907.
- Dulka JG, Stacey NE, Sorensen PW & Van der Kraak GJ 1987 A steroid sex pheromone synchronizes male–female spawning readiness in gold fish. *Nature* **325** 251–253.
- Duntze W, Mackay VL & Manney TR 1970 *Saccharomyces cerevisiae*: a diffusible sex factor. *Science* **168** 1472–1473.
- Duve H & Thorpe A 1981 Gastrin/cholecystokinin (CCK)-like immunoreactive neurones in the brain of the blowfly *Calliphora erythrocephala* (Diptera). *General and Comparative Endocrinology* **43** 381–391.
- Duve H, Thorpe A & Lazarus NR 1979 Isolation of material displaying insulin-like immunological and biological activity from the brain of the blowfly, *Calliphora vomitoria*. *Biochemical Journal* **184** 221–227.
- Eisenschlos CD, Paladini AA, Vedia LMY & Torres HN 1986 Evidence for the existence of an N_G -type regulatory protein in *Trypanosoma cruzi* membranes. *Biochemical Journal* **237** 913–917.
- El Mofty MM & Smyth JD 1964 Endocrine control of encystation in *Opalina ranarum* parasitic in *Rana temporaria*. *Experimental Parasitology* **15** 185–199.
- Escriva H, Safi R, Hanni C, Langlois MC, Saumitou-Laprade P, Stehelin D, Capron A, Pierce R & Laudet V 1997 Ligand binding was acquired during evolution of nuclear receptors. *Proceedings of the National Academy of Sciences of the USA* **94** 6803–6808.
- Falkmer S, Elde RP, Hellerstrom C & Peterson B 1978 Phylogenetics aspects of somatostatin in the gastroenteropancreatic (GEP) endocrine system. *Metabolism* **27** 1193–1196.
- Feldman D, Do Y, Burshell A, Stathis P & Loose DS 1982 An estrogen-binding protein endogenous ligand in *Saccharomyces cerevisiae*: possible hormone–receptor system. *Science* **218** 297–298.
- Feldman D, Tokes LG, Stathis PA, Miller SC, Kurz W & Harvey D 1984 Identification of 17β -estradiol as the estrogenic substance in *Saccharomyces cerevisiae*. *Proceedings of the National Academy of Sciences of the USA* **81** 4722–4726.
- Ferlund P 1974 Structure of red-pigment concentrating-hormone of the shrimp *Pandalus borealis*. *Biochimica et Biophysica Acta* **371** 304–311.
- Firtel RA, Van Haastert PJM, Kimmel AR & Devreotes PN 1989 G-protein linked signal transduction pathways in development: *Dictyostelium* as an experimental system. *Cell* **58** 235–239.
- Fisher FM Jr & Sandborn RC 1962 Production of insect juvenile hormone by the microsporidian parasite *Nosema*. *Nature* **194** 1193.
- Franke RR, Konig B, Sakmar TP, Khorana HG & Hofmann KP 1990 Rhodopsin mutants that bind but fail to activate transducin. *Science* **250** 123–125.
- Gerish G 1987 Cyclic AMP and other signals controlling cell development and differentiation in *Dictyostelium*. *Annual Review of Biochemistry* **56** 853–879.
- Geuns JMC 1978 Steroid hormones and plant growth and development. *Phytochemistry* **17** 1–14.
- Gilbert LI & Schneiderman HA 1958 Occurrence of substrates with juvenile hormone activity in adrenal cortex of vertebrates. *Science* **128** 844.
- Gould SJ & Vrba ES 1982 Exaptation, a missing term in the science of form. *Paleobiology* **8** 4–15.
- Grimm-Jorgensen 1983 Immunoreactive somatostatin in two pulmonate gastropods. *General and Comparative Endocrinology* **49** 108–114.
- Grosman MI 1979 Chemical messengers: a view from the gut. *Federal Procurement* **38** 2341–2343.
- Gustavson AR, Dawson ME & Bonnet DG 1987 Androstenol, a putative human pheromone, affects human (*Homo sapiens*) male choice performance. *Journal of Comparative Psychology* **101** 210–211.
- Hales CN 1985 Lysosomes and prohormone activation. *Nature* **314** 20.
- Hanaoka K & Takahashi SY 1976 Effect of a hyperglycemic factor on hemolymph trealose and fat body carbohydrates in the American cockroach. *Insect Biochemistry* **6** 621–625.
- Henderson IW 1997 Endocrinology of the vertebrates. In *Handbook of Physiology*, vol I, section 13, pp 623–749. Ed WH Dantzler. New York and Oxford: Oxford University Press and American Physiological Society.
- Highnam KC & Hill L 1977 Endocrine mechanisms in the Insecta - I. In *The Comparative Endocrinology of Invertebrates*, pp 104–130. Eds EJW Barrington & AJ Willis. London: Edward Arnold.
- Hwang PK & Fletterick RJ 1986 Convergent and divergent evolution of regulatory sites in eukaryotic phosphorylases. *Nature* **324** 80–84.
- Jackson JMD 1981 Abundance of immunoreactive thyrotropin-releasing hormone-like material in the alfalfa plant. *Endocrinology* **108** 344–346.
- Janakidevi K, Dewey VC & Kidder GW 1966 The biosynthesis of catecholamines in two genera of protozoa. *Journal of Biological Chemistry* **241** 2576–2578.
- Judy KJ, Schooley DA, Hall MS, Bergot BJ & Siddall JB 1973 Chemical structure and absolute configuration of a juvenile hormone from grasshopper corpora allata *in vitro*. *Life Sciences* **13** 1511–1516.
- Kaback DB 1985 Hormone receptor–effector complex evolution. *Nature* **316** 490.
- Kikuyama S, Toyoda F, Ohmiya Y, Matsuda K, Tanaka S & Hayashi H 1995 Sodefrin: a female-attracting peptide pheromone in newt cloacal glands. *Science* **267** 1643–1645.
- King K, Dohlman HG, Thorner J, Caron MG & Lefkowitz RJ 1990 Control of yeast mating signal transduction by a mammalian β_2 -adrenergic receptor and G_s α subunit. *Science* **250** 121–123.
- Kochert G 1978 Sexual pheromones in algae and fungi. *Annual Review of Plant Physiology* **29** 461–486.
- Kurosky A, Markel DE, Peterson JW & Fitch WM 1977 Primary structure of cholera toxin β -chain: a glycoprotein hormone analog? *Science* **195** 299–301.
- Lee J, Horuk R, Rice GC, Bennett GL, Camerato T & Wood WI 1992 Characterization of two high affinity human interleukin-8 receptors. *Journal of Biological Chemistry* **267** 16283–16287.
- Legros F, Videnhoef P, Dumont J, Hanson B, Jeanmart J, Massant B & Conard V 1975 Specific binding of insulin to the unicellular alga *Acetabularia mediterranea*. *Protoplasma* **86** 119–137.
- Leichtling BH, Coffman DS, Yearer ES & Rickenberg HV 1981 Occurrence of the adenylate cyclase 'G-protein' in membranes of *Dictyostelium discoideum*. *Biochemical and Biophysical Research Communications* **102** 1187–1195.
- Lenhoff HM 1968 Behavior, hormones and *Hydra*. *Science* **161** 434–442.

- Le Roith D, Shiloach J, Roth J & Lesniak MA 1980 Evolutionary origin of vertebrate hormones: substances similar to mammalian insulins are native to unicellular eukaryotes. *Proceedings of the National Academy of Sciences of the USA* **77** 6184–6188.
- Le Roith D, Shiloach J, Roth J & Lesniak MA 1981a Insulin: a closely related molecule is native to *Escherichia coli*. *Journal of Biological Chemistry* **256** 6533–6536.
- Le Roith D, Lesniak MA & Roth J 1981b Insulin in insects and annelids. *Diabetes* **30** 70–76.
- Le Roith D, Shiloach J, Berelowitz M, Frohman LA, Liotta AS, Krieger DT & Roth J 1983 Are messenger molecules in microbes the ancestors of the vertebrate hormones and tissue factor? *Federal Proceedings* **42** 2602–2607.
- Le Roith D, Berelowitz M, Pickens W, Crosby LK & Shiloach J 1985a Somatostatin-related material in *E. coli*: evidence for molecular forms. *Biochimica et Biophysica Acta* **838** 335–340.
- Le Roith D, Pickens W, Wilson GL, Miller B, Berelowitz M, Vinik AI, Collier E & Cleland CF 1985b Somatostatin-like material is native to flowering plants. *Endocrinology* **117** 2093–2097.
- Le Roith D, Delahuney G, Wilson GL, Roberts CT Jr, Shemer J, Hart C, Lesniak MA, Shiloach J & Roth J 1986 Evolutionary aspects of the endocrine and nervous systems. *Recent Progress in Hormone Research* **42** 549–587.
- Loose DS & Feldman D 1982 Characterization of a unique corticosterone-binding protein in *Candida albicans*. *Journal of Biological Chemistry* **257** 4925–4930.
- Loose DS, Schurman DJ & Feldman D 1981 A corticosterone binding protein and endogenous ligand in *C. albicans* indicating a possible steroid-receptor system. *Nature* **293** 477–479.
- Loose DS, Price-Stover E, Restrepo A, Stevens DA & Feldman 1983 Estradiol binds to a receptor-like cytosol binding protein and initiates a biological response in *Paracoccidioides brasiliensis*. *Proceedings of the National Academy of Sciences of the USA* **80** 7659–7663.
- Loumaye E, Thorner J & Catt KJ 1982 Yeast mating pheromone activates mammalian gonadotrophs: evolutionary conservation of a reproductive hormone? *Science* **218** 1323–1325.
- Macchia U, Bates RW & Pastan I 1967 Purification and properties of a thyroid-stimulating factor isolated from *Clostridium perfringens*. *Journal of Biological Chemistry* **242** 3726–3730.
- Machlis L, Nutting WH & Rapoport H 1968 The structure of sirenin. *Journal of the American Chemical Society* **90** 1674–1676.
- Mackman RS & Sutherland EQ 1965 Adenosine 3',5'-phosphate in *Escherichia coli*. *Journal of Biological Chemistry* **240** 1309–1311.
- McMorris TC, Seshadri R, Wrihe GR, Aresenault GR & Barksdale AW 1975 Structures of ogoniol-1, -2 and -3, steroid sex hormones of the water mold *Achlya*. *Journal of the American Chemical Society* **97** 2544–2545.
- Manaranche R & L'Hermite P 1973 Etude des amines biogenes de *Glycera convoluta* K (Annelid Polychete). *Zeitschrift für Zelforschung* **137** 21–36.
- Maruo T, Cohen H, Segal SJ & Koide SS 1979 Production of choriogonadotropin-like factor by a microorganism. *Proceedings of the National Academy of Sciences of the USA* **76** 6622–6626.
- Mason RT, Fales HM, Jones TH, Pannell LK, Chinn JW & Crews D 1989 Sex pheromones in snakes. *Science* **245** 290–293.
- Matsumara K 1995 Tetrodotoxin as a pheromone. *Nature* **378** 563–564.
- Melrose DR, Reed HCB & Patterson RLS 1971 Androgen steroids associated with boar odour as an aid to the detection of oestrus in pig artificial insemination. *British Veterinary Journal* **107** 497–501.
- Meselson MS & Radding CM 1975 A general model for genetic recombination. *Proceedings of the National Academy of Sciences of the USA* **72** 358–361.
- Monsma FJ, McVittie LD, Gerfen CR, Mahan LC & Sibley DR 1989 Multiple D₂ dopamine receptors produced by alternative RNA splicing. *Nature* **342** 926–929.
- Mordue W & Stone JV 1976 Comparison of the biological activities of an insect and a crustacean neurohormone that are structurally similar. *Nature* **264** 287–289.
- Mordue W & Stone JV 1977 Relative potencies of locust adipokinetic hormone and prawn red pigment concentrating hormone in insect and crustacean systems. *General and Comparative Endocrinology* **33** 103–108.
- Morley JE, Meyer N, Pekary AE, Melmed, Carlso HE, Briggs JE & Herskam JM 1980 A prolactin inhibitory factor with immunocharacteristics similar to thyrotropin releasing factor (TRH) is present in rat pituitary tumors (GH3 and W5), testicular tissue and a plant material, alfalfa. *Biochemical and Biophysical Research Communications* **96** 47–53.
- Muggeo M, Ginsberg BH, Roth J, Neville DM Jr, Demeyts P & Kahn CR 1979 The insulin receptor in vertebrates is functionally more conserved during evolution than insulin itself. *Endocrinology* **104** 1393–1402.
- Muller DG & Jaenicke L 1973 Fucoserraten, the female sex attractant of *Fucus serratus* L. *FEBS Letters* **30** 137–139.
- Muller DG, Jaenicke L, Donike M & Akintobi T 1971 Sex attractant in brown algae: chemical structure. *Science* **171** 815–817.
- Namba T, Sugimoto Y, Negishi M, Irie A, Ushikubi F, Kakizuka A, Ito S, Ichikawa A & Narumiya S 1983 Alternative splicing of C-terminal tail of prostaglandin E receptor subtype EP3 determines G-protein specificity. *Nature* **306** 32–36.
- Nes W & McKean ML 1977 *Biochemistry of Steroids and other Isopentenoids*. Baltimore: University Park Press.
- Niall HD 1982 The evolution of peptide hormones. *Annual Review of Physiology* **44** 615–624.
- O'Malley BW 1989 Editorial: Did eukaryotic steroid receptors evolve from intracrine gene regulators? *Endocrinology* **125** 1119–1120.
- Paech K, Webb P, Kuiper GGJM, Nilsson S, Gustafsson JA, Kushner PJ & Scanlan TS 1997 Differential ligand activation of estrogen ER α and ER β at AP1 sites. *Science* **277** 1508–1510.
- Pfeffer S & Ullrich A 1985 Epidermal growth factor: is the precursor a receptor? *Nature* **313** 184.
- Plisetskaya E, Kazarov VK, Slititskaya L & Leibson LG 1978 Insulin producing cells in the gut of freshwater bivalve molluscs *Anodonta cygnea* and *Unio pycatorum* and the role of insulin in the regulation of their carbohydrate metabolism. *General and Comparative Endocrinology* **35** 133–145.
- Pullen RA, Lindsay DG, Wood SP, Tickle IJ, Blundell TL, Wollmer A, Krail G, Brandenburg D, Zahn H, Glemann J & Gammeltoft S 1976 Receptor-binding region of insulin. *Nature* **259** 369–373.
- Rao CV & Chegini N 1983 Nuclear receptors for gonadotropins and prostaglandins. In *Evolution of Hormone Receptor Systems*, pp 413–423. New York: AR Liss.
- Richert ND & Ryan RJ 1977 Specific gonadotropin binding to *Pseudomonas maltophilia*. *Proceedings of the National Academy of Sciences of the USA* **74** 878–882.
- Riedel H, Dull TJ, Schlessinger J & Ullrich A 1986 A chimaeric receptor allows insulin to stimulate tyrosine kinase

- activity of epidermal growth factor receptor. *Nature* **324** 68–70.
- Rosenzweig JL, Havrankova J, Lesniak MA, Brownstein M & Roth J 1980 Insulin is ubiquitous in extrapancreatic tissues of rat and humans. *Proceedings of the National Academy of Sciences of the USA* **77** 572–576.
- Saden-Krekula M, Tajic M & Kolbah D 1971 Testosterone, epitestosterone and androstenedione in the pollen of Scots pine *P. silvestris*. *Experientia* **27** 108–109.
- Saxe CL, Johnson RL, Devreotes PN & Kimmel AR 1991 Expression of cAMP receptor-gene of *Dictyostelium* and evidence for a multigene family. *Genes and Development* **5** 1–8.
- Schapp P 1984 cAMP pulses coordinate morphogenetic movement during fruiting body formation of *Dictyostelium minutum*. *Proceedings of the National Academy of Sciences of the USA* **81** 2122–2126.
- Schar G, Price-Stover E, Clemons KV, Feldman D & Stevens DA 1986 Progesterone binding and inhibition of growth in *Trichophyton mentagrophytes*. *Infection and Immunity* **52** 763–767.
- Schildknecht H, Siewerdt R & Maschwitz 1966 A vertebrate hormone as defensive substance of the water beetle *Dytiscus marginalis*. *Angewandte Chemie International Edition (English)* **5** 421.
- Schildknecht H, Birringer H & Maschwitz U 1967 Testosterone as protective agent of the water beetle *Ilybius*. *Angewandte Chemie International Edition (English)* **6** 558.
- Schinz A, Hinsh KD & Hildebrand E 1989 Enzymatic and immunological detection of a G-protein in *Halobacterium halobium*. *FEBS Letters* **249** 59–61.
- Schwabe C, Le Roith D, Thompson RP, Shiloach J & Roth J 1983 Relaxin extracted from protozoa (*Tetrahymena pyriformis*): molecular and immunologic properties. *Journal of Biological Chemistry* **258** 2778–2781.
- Schwemmler W 1984 Evolution of the precytes. In *Reconstruction of Cell Evolution: a Periodic System*, pp 45–89. Ed W Schwemmler. Florida: CRC Press.
- Shemshedini L & Wilson TG 1990 Resistance to juvenile hormone and an insect growth regulator in *Drosophila* is associated with an altered cytosolic juvenile hormone binding protein. *Proceedings of the National Academy of Sciences of the USA* **87** 2072–2076.
- Shimomura O, Suthers HLB & Bonner JT 1982 Chemical identity of the acrasin of the cellular slime mold *Polysphondylium violaceum*. *Proceedings of the National Academy of Sciences of the USA* **79** 7380–7384.
- Slama K & Williams CM 1965 Juvenile hormone activity for the bug *Pyrrochoris apterus*. *Proceedings of the National Academy of Sciences of the USA* **54** 411–414.
- Starr RC & Jaenicke L 1974 Purification and characterization of the hormone initiating sexual morphogenesis in *Volvox carteri* f. *magariensis*. *Proceedings of the National Academy of Sciences of the USA* **71** 1050–1054.
- Stoka AM 1987 Ecdysteroids, juvenile hormones and metamorphosis in *Triatominae*. In *Chagas' Disease Vectors*, vol III, pp 71–99. Eds RR Brenner & AM Stoka. Florida: CRC Press.
- Stoka AM 1996 Activity of juvenile hormone and juvenile hormone analogues on the growth of *Trypanosoma cruzi*. *Journal of Steroid Biochemistry and Molecular Biology* **59** 495–500.
- Stoka AM, Ronderos JR & Reboredo GR 1987a Insect control by endocrine disorder. In *Chagas' Disease Vectors*, vol III, pp 139–147. Eds RR Brenner & AM Stoka. Florida: CRC Press.
- Stoka AM, Salomon OD & Noriega FG 1987b Physiology of *Triatominae*'s reproduction. In *Chagas' Disease Vectors*, vol II, pp 109–129. Eds RR Brenner & AM Stoka. Florida: CRC Press.
- Stoka AM, Rivas C, Segura EL, Rodriguez JB & Gros EG 1990 Biological activity of synthetic juvenile hormone analogues (JHA) for *Trypanosoma cruzi*. *Zeitschrift für Naturforschung* **45b** 96–98.
- Stone JV, Mordue W, Batley KE & Morris HR 1976 Structure of locust adipokinetic hormone, a neurohormone that regulates lipid utilization during flight. *Nature* **263** 207–211.
- Sturaro A, Guerriero A, Declausen R & Pietra F 1982 A new unexpected marine source of a moulting hormone; isolation of ecdysterone in large amounts from the zoanthid *Gerardia savaglia*. *Experientia* **38** 1184–1185.
- Tainer JA, Getzoff ED, Alexander H, Houghten RA, Olson AJ & Lerner RA 1984 The reactivity of anti-peptide antibodies is a function of the atomic mobility of sites in a protein. *Nature* **312** 127–134.
- Tata JR 1964 Basal metabolic rate and thyroid hormones. *Advances in Metabolic Disorders* **1** 153–189.
- Thambi NC, Quan F, Wolfgang WJ, Spiegel A & Forte M 1989 Immunological and molecular characterization of G_o-like proteins in the *Drosophila* central nervous system. *Journal of Biological Chemistry* **264** 18552–18560.
- Toms RB 1984 Environmental selective pressure and the origin of insect metamorphosis. *Journal of the Entomological Society of South Africa* **47** 269–276.
- Vallesi A, Giuli G, Bradshaw RA & Luporini P 1995 Autocrine mitogenic activity of pheromones produced by the protozoan ciliate *Euplotes raikovi*. *Nature* **376** 522–524.
- Van Haastert PJM, De Wit RJW, Grijma Y & Konijn TM 1982 Identification of a pterin as the acrasin of the cellular slime mold *Dictyostelium lacteum*. *Proceedings of the National Academy of Sciences of the USA* **79** 6270–6274.
- Vladusic EA, Bussmann LE, Visconti PE, Stoka AM, Rodriguez JB, Gros EG & Charreau EH 1994 Effects of juvenile hormone on mammalian steroidogenesis. *Journal of Steroid Biochemistry and Molecular Biology* **50** 181–187.
- Watanabe M & Po L 1976 Membrane bound 3 β - and 17 β -hydroxysteroid dehydrogenase and its role in steroid transport in membrane vesicles of *Pseudomonas testosteroni*. *Journal of Steroid Biochemistry* **7** 171–175.
- Watanabe M, Phillips K & Chen T 1973a Steroid receptor in *Pseudomonas testosteroni* released by osmotic shock. *Journal of Steroid Biochemistry* **4** 613–622.
- Watanabe M, Phillips K & Watanabe H 1973b Induction of steroid-binding activity in *Pseudomonas testosteroni*. *Journal of Steroid Biochemistry* **4** 623–632.
- Weber IT, Takio K, Titani K & Steitz M 1982 The cAMP-binding domains of the regulatory subunit of cAMP-dependent protein-kinase and the catabolite gene activator are homologous. *Proceedings of the National Academy of Sciences of the USA* **79** 7679–7683.
- Wedemayer GJ, Patten PA, Wang LH, Schultz PG & Stevens KC 1997 Structural insights into the evolution of an antibody combining site. *Science* **276** 1665–1669.
- Weinberger C, Thompson CL, Ong ES, Lebo R, Gruol DJ & Evans RM 1986 The c-erb-A gene encodes a thyroid hormone receptor. *Nature* **324** 641–646.
- Weiss M, Ingbar SH, Winblad S & Kasper DL 1983 Demonstration of a saturable binding site for thyrotropin in *Yersinia enterocolitica*. *Science* **219** 1331–1333.
- Welsh JH & King EC 1970 Catecholamines in planarians. *Comparative Biochemistry and Physiology* **36** 683–688.
- White BA & Nicoll CS 1979 Prolactin receptors in *Rana catesbeiana* during development and metamorphosis. *Science* **204** 851–853.

- Whiteway M, Hougan L, Dignard D, Thomas DI, Bell L, Saari GC, Grant FJ, O'Hara P & MacKay VL 1989 The STE4 and STE18 gene of yeast encode potential β and γ subunits of the mating factor receptor-coupled G-protein. *Cell* **56** 467–477.
- Wilkinson LE & Pringle JR 1974 Transient G1 arrest of *S. cerevisiae* of mating type α by a factor produced by cells of mating type a . *Experimental Cell Research* **89** 175–187.
- Williams CM, Morhead LV & Pulis JF 1959 Juvenile hormone in thymus, human placenta and other mammalian organs. *Nature* **183** 405.
- Wilson AG, Carlson SS & White TJ 1977 Biochemical evolution. *Annual Review of Biochemistry* **46** 573–639.
- Yang NN, Venugopalan M, Hardikar S & Glasebrook A 1996 Identification of an estrogen response element activated by metabolites of 17 β -estradiol and raloxifene. *Science* **273** 1222–1225.
- Zimmerman EG 1979 Peptides of the brain and gut: introductory remarks. *Federal Proceedings* **38** 2286–2287.
- Zioudrov C, Streaty RA & Klee WA 1979 Opioid peptides derived from food proteins. The exorphins. *Journal of Biological Chemistry* **254** 2446–2450.

REVISED MANUSCRIPT RECEIVED 17 August 1998