

Sleep and its possible role in learning: a phylogenetic view

Stefania Piscopo

Lab. of Animal Physiology and Evolution, Stazione Zoologica Anton Dohrn, 80121 Napoli, Italy

TABLE OF CONTENTS

1. Abstract
2. Introduction
3. Main features of sleep
4. General ideas about the phylogeny of sleep
5. Possible functions of sleep
6. Sleep and synaptic plasticity
7. Cnidarians
8. Nematodes
9. Molluscs
10. Arthropods
11. Discussion & Conclusion
 - 11.1. Towards a general hypothesis
12. Acknowledgements
13. References

1. ABSTRACT

A fascinating but still partly unsolved scientific question is to understand the function/s of sleep. Many hypotheses have been proposed, and in this review I focus on the idea that sleep has a role in supporting brain plasticity, learning and memory. Although the majority of sleep research is concentrated on vertebrates, especially humans and mammals, sleep-like behaviour is also evident in some invertebrates. Though surprisingly there are very few studies that have directly tested if there is a mechanistic link between sleep and plasticity. I argue that a phylogenetic approach gives potential insight into the mechanisms and roles of sleep, by allowing an analysis by comparison of the evolution of sleep and rest/activity cycles in organisms showing radically different lifestyles and brain plans. This review attempts to set the scene for a much more profound analysis of these issues using phylogenetic tools.

2. INTRODUCTION

Sleep in most vertebrates is a heterogeneous state characterized by behavioural, electrophysiological and energetic aspects. In invertebrates, our knowledge of sleep is growing rapidly though we are as yet far from a complete synthesis. At the same time, it is clear that a comparative approach that examines sleep features in phylogeny may help in understanding sleep evolution, and may be useful to indicate what could be the universal functions of this physiological state.

The current list of animal phyla studied from this respect has been rightly, and highly biased, as the main effort has been directed at understanding mammalian sleep. Until recently, there were only a few reports, based mostly on behavioural observations, dedicated to invertebrate “rest” or “sleep-like behaviour”.

Sleep and its possible role in learning

This situation has changed dramatically with the advent of cheap and rapid throughput genome sequencing (“genomics”) which has made it possible to sequence many small invertebrate genomes. This has opened the possibility of using combined functional/genomic approaches to address basic questions. As a result, we now know that a complicated phenomenon such as sleep is also present in a small insect like the fruit-fly. However although this represents a very restricted phylogeny, it has already been suggested that the phylogenetic approach may be a good tool to understand in more depth the evolution and the function of sleep (1,2).

In this review for the first time I have listed the principal studies done on sleep or rest activity cycles in invertebrate phyla and using this information I discuss the important proposal that the function of sleep could be related to synaptic plasticity and learning.

3. MAIN FEATURES OF SLEEP; VERTEBRATES

Before proceeding with a detailed evaluation of sleep and sleep-like processes in invertebrates, it is worth reviewing what we already know about the process in warm blooded vertebrates. Sleep is a heterogeneous functional state characterized by reduced responsiveness and homeostatic regulation. Modern research on sleep started in the 30s when the EEG method was invented (3) and it became possible to record brain electrical activity and to correlate this with behaviour. On the basis of this correlation, it was possible to identify 2 different kinds of sleep: slow wave or synchronised sleep (SWS) and REM or paradoxical sleep (PS). These two sleep states have been shown to be present in both mammals (4) and birds (5). SWS is characterized by low frequency and high amplitude waves, while the ‘paradox’ in PS is the existence of a desynchronised trace consisting of high frequency and low amplitude waves, similar to an awake state. The paradoxical state may be distinguished from the awake state only by correlating it with the loss of muscular tone, rapid eye movements, irregular heart and respiratory rate, variation of the blood pressure and presence of dreams. The sleep cycle itself is strongly influenced by the endogenous clock, but also by external factors such as light and temperature, and is regulated by homeostatic mechanisms. In fact, lack or deprivation of sleep leads to compensation (sleep rebound). Lastly it should be underlined here that the term ‘sleep’ when used in the vertebrate/mammalian context emerges from this strong tradition in behaviour and neurophysiology.

4. GENERAL IDEAS ABOUT THE PHYLOGENY OF SLEEP

Explanations of sleep function have tended to reflect our focus on mammals. It has been stated variously that sleep evolved with brain complexity, homeothermy or increasing system complexity. For example, it has been claimed that synchronized sleep was present in the Palaeozoic era, while REM sleep origin was more recent, occurring along with homeothermy (6). Until recently PS was found only in birds and mammals and was present in

a intermediate state in echidna, a monotreme which represents a kind of primitive mammal (7). Recently it has been shown that echidna sleep has a REM component and hence a PS state (8) as well as the platypus, another member of the monotremes (9). While, the idea that sleep evolved along with brain complexity, learning and memory and intelligence is somewhat undermined by the spectacular lack of PS in certain key mammalian species such as cetaceans that must sleep with one hemisphere at a time to allow the other hemisphere control respiration. These animals show only slow cortical activity comparable to SS and an absence of PS (10). Equally, in reptiles, (11) amphibians and fish (12) no REM sleep has been found, although for practical reasons it has been difficult to define clearly this characteristic of their sleep. Thus as the general situation in vertebrates is unclear with no apparent unifying pattern, a critical comparison with invertebrates could be instructive. It should be noted first that when we talk of invertebrates, it has only recently been possible to talk about “invertebrate sleep”. In the past the term “rest” has been seen to be a more appropriate term.

Historically, research on invertebrate sleep started last century with behavioural descriptions, but since then, it has been possible to make correlative electrical recordings and now molecular and now genetic analysis as well. In Figure 1 I show a simplified scheme of the phylogenetic tree of the animal kingdom, indicating phyla where sleep has been shown to exist (red spot). The simplest animals (in terms of brain complexity) where such a state has been described are the “cubomedusae” or box jellyfish that are different from other cnidarians as they have a very specialised visual system that allows them to perform visually guided avoidance (13). Other simple animals include the nematode worm *C. elegans*, (14) an invertebrate with a very simple ‘brain’ consisting of only 302 neurons. Looking into the ‘upper’ part of the tree, one sees that in the area of the celomatic protostomes, that sleep has been discovered in 2 other phyla (molluscs and arthropods). Among molluscs, *Octopus vulgaris* is a cephalopod whose outstanding ability to learn has been described in detail (15,16) and here it has been possible to make an *in vivo* electrical recording of sleep in a free moving animal (17). Arthropods have been the most studied phylum in invertebrate sleep research, as the first observations of this sort were carried out on scorpions and cockroaches (18,19,20) and recently *Drosophila melanogaster* has become a key model in this field of research (21).

5. POSSIBLE FUNCTIONS OF SLEEP

As stated above, the function/s of sleep have/s always been and remain an open question despite the many different hypotheses proposed. Some authors consider sleep to be a restorative period when all cell synthesis and metabolic operations occur. For example, Adam and Oswald proposed that during SS there is an intense cell proliferation and tissue restoration, considering also that the presence of growth hormone in the blood is higher during the first hours of sleep (22). The same is true for sex hormones and prolactin (23), while in other experiments it

Sleep and its possible role in learning

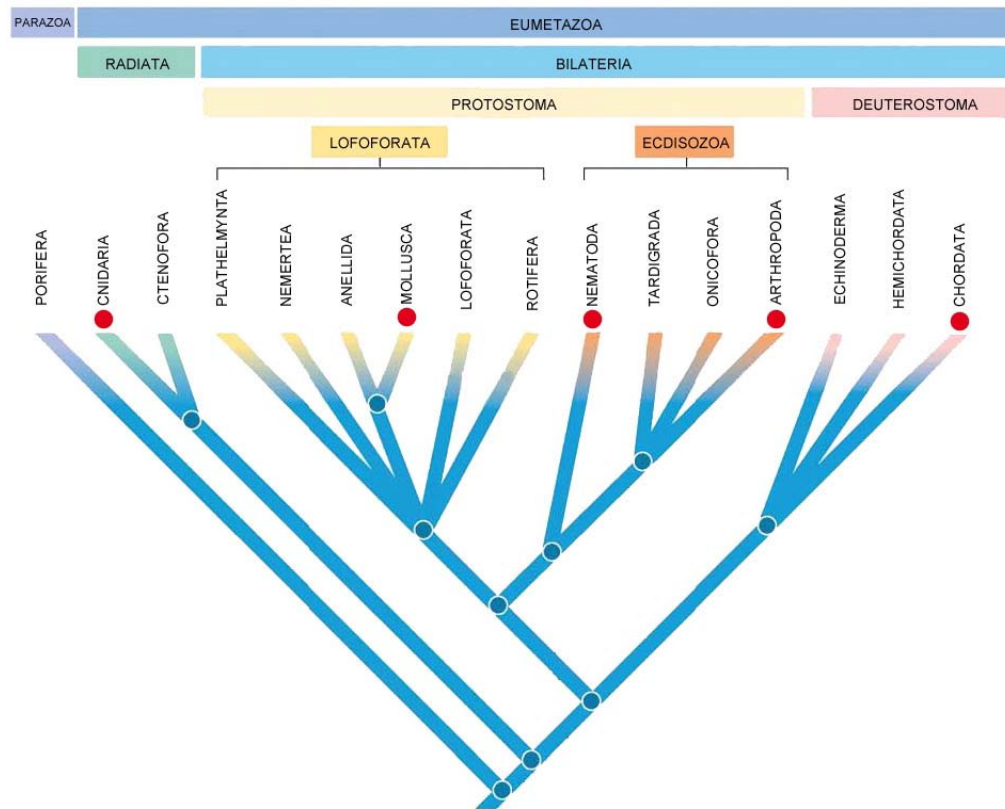


Figure 1. Animal phylogenetic tree showing major transitions and the position of key species covered in this review.

has been seen that during SS there is a decrease in protein synthesis in the brain (24). In another theory, based on studies on mammalian basal metabolism, which is reduced by the 10 % during SS, SS was proposed to be a stage for energy conservation (25,26). This hypothesis may be appropriate for the animals that in difficult conditions (like lack of food), need to save energy. If sleep represents a stage of energy saving, the animal could use it to have a better possibility of survival. To support this hypothesis, there is the consideration that adaptive stages like torpor or hibernation start from SS and not from the awake state. So SS could be the first step of a series of different energy saving levels. Other hypotheses claim that during sleeping other necessary operations such as thermoregulation occur in the brain (27), or detoxification (28).

Many authors propose that sleep facilitates brain plasticity to favour the adaptation of behaviour to any situation. In this way the brain would not be constrained to follow only genetic programs, but would be able to change its structure and function, and respond to environmental changes (29,30). According to this work, since the brain tends to be more complex in more complex animals, where it is necessary to retain large amounts of new information together with 'pre-programmed' instinctive functions (6,31), the information could be best reconciled in the absence of other (confounding) major input. This operation would be best done during sleep when the animal is isolated from the environment and the brain is not

involved in other active operations. The relationship between sleep and memory was first noticed at the end of the nineteenth century (32) and by many authors it is still considered a valid if largely untested hypothesis for the function of sleep.

6. SLEEP AND SYNAPTIC PLASTICITY

As intimated in the last section, the involvement of sleep in memory is an attractive explanation for sleep function, thus any changes should occur through alterations in synaptic plasticity the cellular and synaptic correlate of the memory trace. At present there are two main views, the "ontogenetic hypothesis" and the "memory consolidation hypothesis". According to the ontogenetic hypothesis sleep has an important role in brain maturation, as the amount of REM sleep decreases with age and is maximum in neonatal life (33). Many studies have been done whose results support this idea. For instance some authors believe that REM sleep is fundamental for brain growth, connectivity and synaptic plasticity (34) and others underline the role of sleep in the development of central visual pathways (35). Before REM sleep was discovered (4), the idea that sleep was involved in learning and memory process was already present, in fact in 1885 Ebbinghaus, a German psychologist that was carrying experiments on memory, discovered that the amount of forgetting was reduced by sleep, but he rejected

Sleep and its possible role in learning

his data thinking that he had made accidental errors in his experiments (32).

Jenkins and Dallenbach, 50 years later, studying his work, reconsidered his data and started making experiments on human subjects. They discovered that people who slept after reading a list of nonsense phrases, could remember better. With these data they proposed an *interference theory of forgetting*, in which they assumed that since during the night the brain rests, the memory persists because there is less interference (36).

After the discovery of REM sleep, several others hypotheses were proposed about the relationship between sleep and memory (37), among these, there are some authors who believe that the amount of REM sleep is important for memory consolidation (38,39,40,41,6,42,43) while others that believe the REM sleep does not have an important role in the elaboration of memories (44,45). After a series of experiments on human subjects, it has been proposed that sleep has more than one role in the elaboration of memory traces, since it could be useful to consolidate perceptual and motor skill learning, simple declarative memory, emotional memories and complex cognitive learning (46).

Using non invasive neuroimaging techniques such as PET (positron emission tomography) and fMRI (functional magnetic resonance imaging), it has been possible to correlate learning behaviour while awake with neuronal activity during sleep and it has been shown that entire macroscopic cortico-subcortical networks change depending on experience during the previous awake state. Moreover with experiments of sleep deprivation, there was a lack of recent memories as a consequence (47). Since it has been postulated as a result of experiments at the behavioural, physiological, cellular and molecular levels, that synaptic plasticity processes are altered when sleep is disturbed, it has been proposed that the primary function of sleep might be the facilitation of synaptic plasticity (48) however a direct connection between the two processes is currently lacking in vertebrates. However vertebrates are not the only animals to have developed complex brains, and synaptic plasticity has been detected in most animal phyla. How does the above hypothesis hold up across animal phylogeny?

7. CNIDARIANS

Cnidarians are some of the simplest metazoan animals capable of independent locomotion and they live in one of the most challenging environments on earth: the sea. In general they do not show much in the way of complex neural processing or behaviour, apart from *Chironex fleckeri*, a cubomedusa known also as the “sea wasp” because of its lethal sting. This jellyfish is notable because even if it has no centralised brain, it has a complex visual system with 24 eyes of 4 different types, each specialised for a different task (unlike other jellyfish). By means of ultrasonic transmitters (biotelemetry) it was possible to follow the animals with an underwater directional microphone and track their activity. The data showed that

jellyfish were more active during the daylight hours (from about 0600 to 1500), when they moved with a speed of about 212 m an hour, while from 1500 to 0600, they moved on average at less than 10 m an hour. During the night, jellyfish were clearly inactive, lying motionless on the sea floor, with tentacles completely relaxed and absence of bell pulsation. This rest was disturbed by external stimuli like strong lights or vibrations that caused the animals to rise from the sea floor and swim around briefly, falling back into an inactive state on the sand soon after. Using this method the author recorded 15h of behavioural sleep (49).

The discovery of a sleep-like state in jellyfish supports the hypothesis of an ancient origin of sleep and show that its basic function is not coupled to brain complexity. An evolutionary analysis was used to suggest that sleep in this animal could be linked to the conflict produced by a huge request of neural elaboration from complex eyes on the multifunctional neuronal circuit. This conflict may be solved if all the “non-urgent process” can be done during sleep. Without sleep, selection would favour the evolution of circuits dedicated to single or few tasks with a corresponding minor efficiency. (50).

8. NEMATODES

Nematodes are very versatile worms that include species adapted to different habitats, they may be free living terrestrial, or aquatic animals or parasites. Perhaps the best understood in this phylum is the free-living worm *C. elegans*, that although ‘simple’, possesses a nervous system that has been studied in detail.

Some recent work on invertebrate sleep has been carried out on *C. elegans*, whose quiescence period (associated with lethargus), shows sleep-like properties of reversibility, reduced responsiveness and homeostasis. This sleep-like behaviour is under homeostatic regulation and with transgenic experiments, it was found that sensory neurons have a role in the regulation of lethargus. Equally it was shown that the genetic control of sleep is a conservative process as there is a common genetic regulation of sleep-like states in arthropods and nematodes. Finally it was shown that in this species the sleep-like state has a role in the growth and development of the nervous system (14).

9. MOLLUSCS

The Molluscs are a very heterogeneous phylum including several different species living in different habitats and among which cephalopods have the most complex brain, with well developed and elaborate sensory and motor organs. Their nervous system are large and consist of ‘nuclear’ specialized lobes to a much greater extent than other molluscs. Unsurprisingly perhaps, their behaviour is similar in many respects to higher vertebrates, a result according to Packard, of their ancient and ongoing competition with fish (51). *Octopus vulgaris* for example, exhibits at least 3 different kinds of complex learning: tactile learning, visual discrimination and observational learning (52), in addition to the simple forms of learning common to most invertebrates. These advanced abilities

Sleep and its possible role in learning

have been associated (mainly with lesion experiments) with the supra oesophageal lobes of the brain and it has been hypothesised that a parallelism exists between the structure and the function of these lobes and the vertebrate hippocampus. Altman noticed that *Octopus vulgaris* is more active during the night (53), and later it was demonstrated that they generally catch prey during the night and during the day prefer to stay in their dens (54). In 1995 Cobb demonstrated the presence of a circadian cycle in another species of octopus (*Eledone cirrhosa*) (55). However, there seems to be no evidence for a circadian influence on visual and tactile discrimination learning (56). Recently, behavioural, and electrical evidence, has provided the first indication that something like the well known “off-line activity” associated with vertebrate sleep and memory processing when sensory input is closed down is also occurring in an invertebrate (17). In fact it has been shown in *O. vulgaris* that rest periods are characterised by a time of day effect, quiet behaviour, decreased responsiveness to external stimuli, behavioural rebound after deprivation, and selective modification of electrical activity in the supra oesophageal lobes (17). As octopus has a complex brain and ‘advanced’ ‘sleep-like behaviour’ the question naturally arises as to whether this indicates that sleep has a role in learning and memory.

10. ARTHROPODS

The first study on activity cycles in arthropods was carried out by Lindauer in 1952, who wrote a detailed paper about the life of bees in a beehive. He followed their activity for 8 days continuously and he was able to see how they organize their work and distribute the tasks inside their community. In particular he made long term observations of the life of single bees and made graphs of their activity with frequency histograms. The results were graphically presented showing different activities; bees cleaning their cells, caring for the larvae, building cells, checking that there was no danger for the beehive, and working for temperature regulation. From a comparison of different activities done during the night and the day, it emerged that, although they never stopped working, the quantity of each category is always larger during the hours of light. Lindauer concluded that he wasn’t able to say if the period of rest occurred in a determined hour of the day or night, but he also said that “*the rest-activity cycle is the same during the light and during the dark*”, talking for the first time of a rest-activity cycle in an arthropod. (57). More recently, honey- bee neural activity during sleep has been measured by recording from the optic lobes, and shows the existence of an endogenous cycle. Sleep is also characterized by movements of the antennae at regular intervals (58) where the posture of the animal including the position of the head and antennal movement indicate the depth of sleep (59). As sleep deprived honey bees compensate the sleep deficit by intensifying the sleep process, it was also proved that sleep in honey bees is controlled by regulatory mechanisms (60). Other arthropods such as scorpions and cockroaches exhibit rest cycles; here there are not only differences in the behaviour, such as the existence of a 24hr variation of vigilance and a reduced responsiveness, but also the

presence of a homeostatic response to rest deprivation, demonstrating the existence of regulatory aspect of sleep (18,19,20). It has been shown that an increased metabolic rate (82%) occurs in sleep deprived cockroaches which eventually die (61). As this animal is ‘cold-blooded’ this data is in contrast with the hypothesis that increased metabolic heat production is produced by a perturbation of the temperature regulating system, caused by the deprivation of sleep (62) and supports the idea that insects need sleep like mammals (61). Finally another arthropod that shows sleep like state is crayfish, where a quiescent behavioural state exists where the animal tends to lie in one position and shows an elevated threshold for vibratory stimulation in this state. Furthermore this state is accompanied by a distinctive form of slow wave electrical activity of the ‘brain’ ganglia (63,64).

In the last few years, *Drosophila melanogaster* has emerged as key model for modern studies on sleep. This is because many because of the features of its sleep are similar to mammalian sleep and it has the advantage of allowing advanced genetic approaches such as rapid mutagenesis screening. Sleep in the fruit fly was first described in 2000 by two groups working independently ; it is characterized by immobility (5 or more minutes) an increased arousal threshold influenced by homeostasis and a circadian cycle (21,65). Like mammalian sleep, drosophila sleep is regulated by the catabolism of monoamines, is age dependent, is modulated by stimulants and hypnotics, and has several molecular markers in common with mammals (65), Amylase (66), and glycogen in the brain changes in relationship to rest and activity (67). According to Zimmerman, sleep deprivation causes a nervous system stress response that may induce a change in immune response gene expression, that would provoke a down regulation of genes and an encode for proteins that control multiple steps in protein metabolism. This would influence calcium homeostasis and neuronal excitability. So a forced prolonged wakefulness results in a huge waste of energy and this would be stopped by multiple mechanisms instead of accumulation of chemical factors (68). With electrophysiological recording of local field potentials in the medial brain between the mushroom bodies, it has also been demonstrated that during sleep there are changes in brain activity, (69). Moreover chemical ablation of the mushroom bodies provoked reduced sleep, demonstrating that they have an important role in sleep regulation (70). Mutagenesis studies have been done on *Drosophila* and 5000 mutants have been screened. From these data it emerged that, although the presence of 10 short sleeper lines and 4 lines with no compensation after sleep deprivation, the homeostatic regulation of sleep are highly conserved phenotypes (71,72). At a molecular level, circadian and homeostatic mechanism of sleep regulation interacts and the time of the day has an influence on the expression of many wakefulness-related and sleep-related *Drosophila* transcripts (73,74). Another study was done on the neurobiological mechanisms of arousal in the *Drosophila* brain, which is modulated and regulated by dopamine (75,76,77). Bushey in 2007 identified a gene, Hk, which is necessary to maintain normal sleep, and provided genetic evidence that short sleep and poor

Sleep and its possible role in learning


















Behaviour body immobility	Behaviour increased arousal threshold	Behaviour & homeostasy Compensation after deprivation	Electrical recording	Metabolic changes	Alterations in Gene expression	Sleep mutants
						
						
						
						

Figure 2. Scheme of experiments done on invertebrates investigating the presence of sleep.

memory are linked (78). Probably the most important results of this work in relation to the function is sleep is the finding that wakefulness leads to up regulation of the expression of three categories of genes; energy metabolism, cellular stress related and activity dependent synaptic potentiation. During sleep, genes in the categories, synaptic downscaling, synthesis and maintenance of membranes and lipid metabolism are up-regulated and the hypothesis has emerged that sleep may have a major function in controlling synaptic weight (79). Recently it has been shown that there is a connection between social experience and sleep in flies, in fact socially enriched individuals sleep more than socially impoverished ones and if both the types were sleep deprived for 24 h, they compensated the sleep loss by going back to their previous levels. Moreover sleep mutants (clock, timeless and cycle genes) have disrupted circadian rhythms and normal experience dependent responses in sleep need. It seems also that experience dependent changes in sleep are modulated by dopamine. In an elegant series of experiments it was found that in courtship conditioning of male flies, which induces long-term memory, sleep time was significantly increased and conversely when this was blocked by sleep deprivation for 4 hours after conditioning both training induced changes in sleep bout and courtship memory were abolished. (80). The results begin to illustrate a strong link between learning and memory and sleep function, the reader is referred to several recent reviews that discuss 'fly-sleep' in depth (79,81,82).

11. DISCUSSION & CONCLUSIONS

A recent paper reported the discovery of a dinosaur from the cretaceous that was found in an avian-like sleeping posture, (83) gives another clue about how sleep has had a long and important history. Phylogeny

however points to a more ancient origin of sleep, as we now know it is present in simple animals like Cnidarians.

In Figure 2 I show a simple scheme of the experiments done on invertebrate sleep. The criteria applied in investigation of sleep are listed in the upper part of the scheme, three are behavioural and consider posture, responsiveness to stimuli and sleep rebound that imply homeostatic regulation, and the fourth is electrical recording which provides a useful indication of what happens in the brain while the animal is sleeping. Finally, there are methods to check if metabolic changes occur and if genetic techniques may be applied (such as the search for sleep mutants and if there are effects produced by the alteration of gene expression) and this approach in arthropods has already yielded important results that have been shown to have universal (in terms of phylogeny) application. What are the possible new avenues one might explore with the phylogenetic approach? Starting from less complicated organisms, the work done on "*cubomedusae*" is purely based on behavioural data, since the author followed animal movements for 24 hours with a telemetric transmitter. So although from this kind of analysis was possible to see that the speed of the animal was higher during the day, while in the night was much lower, indicating that the medusa was likely to be resting. No other evidence has been presented, though and considering the simplicity of the nervous system it would be useful to test the hypothesis that 'off' line processing occurred during sleep by making electrical recordings from the neural networks in the swimming animal. In addition, a critical approach to understand the circuitry may be revealing, as for example, a lack of motor activity during visual processing would require a damping circuit (analogous to the inhibition by the reticular activating system in mammals) that actively suppresses motor output, a situation that is difficult to imagine in a simple circuit such as this.

Sleep and its possible role in learning

Table 1. The brains of invertebrates, discussed in the text, and their behavioural capacity compared

PHYLUM	PLASTIC AREA IN THE BRAIN	BEHAVIOUR
CNIDARIA (<i>Chironex fleckery</i> , <i>Tripedalia cystophora</i> , <i>Chiropsella Bronzie</i>)	Complex visual system	Fish-like behaviour Visually guided avoidances
NEMATODA (<i>Caenorhabditis elegans</i>)	Neural circuit with interneurons RIA (integration and processing)	Complex learning Plastic behaviour
MOLLUSCA (<i>Octopus vulgaris</i>)	Vertical lobe (LTP)	Complex learning Plastic behaviour
ARTROPODA (<i>Drosophila melanogaster</i> , <i>Apis mellifera</i> , <i>Procambarus clarkii</i>)	Mushroom bodies (insect) Visual interneurons network (crayfish)	Complex learning Plastic behaviour

In nematodes, recent work shows that they also possess sleep-like behaviour and have the advantage of providing another genomic model to compare with that of *Drosophila*. The only weak point is the difficulty in possibility of carrying out correlations between behaviour and 'brain' activity (though it is notable that it is likely that this very simple animal sleeps).

In molluscs the work on cephalopods lacks the support of genetic information, and this should be an area of great interest to explore in the future especially considering the possible tie-in with complex behaviour and sleep.

Arthropods are undoubtedly the invertebrate phylum where the majority of the 'phylogenetic' work on sleep has been concentrated. Apart from the basic observation of posture and lack of responsiveness, in scorpions, cockroaches, honey bees and *Drosophila*, it has been possible to also examine also if there is a sleep rebound after deprivation. In addition metabolic changes have also been noticed in cockroaches and *drosophila*. In honeybees, *drosophila* and crayfish brain electrical recording has also been carried out. It's clear that, at the moment the studies on *Drosophila* are the most complete and the fruit fly has become a key model making a major contribution to understanding the function of sleep.

11.1. Towards a general hypothesis

The hypothesis that has emerged from comparative work so far suggests that synaptic weight (a major regulator of synaptic plasticity) may be a phylogenetically conserved mechanism in sleep/wake cycles. It is then profitable to examine the potential correlates/way forward in other more phylogenetically remote species. In Table 1 I summarise the presence of specific 'plastic' areas of the brain of invertebrates that exhibit sleep and review both their 'behaviour' and capacity for learning. The 'simplest' animals in the list are box jellyfish although they have a sophisticated visual system which allows them to filter information earlier than a more general visual system. Their eyes are in fact camera-type eyes morphologically similar to the vertebrate and cephalopod eyes. (84). Moreover cubomedusae exhibit 'fish-like' behaviour, showing strong directional swimming combined with rapid turns (up to 180° in two bell contraction) (13), positive phototaxis (85,86) and are able to perform visually guided obstacle avoidance (13). As many hypotheses involve sleep during neural development, and it seems that REM sleep is important for the correct development of the rudimentary neural circuitry in the

visual system in vertebrates (87), sleep might be fundamental for each animal that needs a correct development of its eyes. It's interesting to underline the fact here that sleep has been found only in jellyfish with complex eyes. In *C. elegans* it has been possible to understand the molecular and cellular basis of neural plasticity. In a recent experiment in fact, thermo-taxis behaviour of the worm was analyzed at molecular, neuronal and neural circuit level and has been proposed that the work of integrating and processing learned information is done by interneuron RIA, that represents an area of synaptic plasticity in the brain of the nematode (88). In the complex brain of *Octopus vulgaris* the vertical lobe is well known to be prevalently involved in memory processing. As Brown managed to record an active electrical trace on the vertical lobe of a sleeping octopus, it may not be unreasonable to conclude that its activity during quiet behaviour reflects some form of memory processing. The rest state is characterized by a 'body off/brain-different' situation as its electrical trace is active while eyelids are closed and its posture is relaxed. Moreover there is also a sleep rebound after deprivation showing homeostatic regulation. *Octopus vulgaris* is a cephalopod that is able to perform associative learning, observational learning (52), and in addition exhibits both short and long term memory (15,16) under laboratory conditions. Thus octopus brain is arguably the most sophisticated of all invertebrates and has been used not only for learning and memory experiments but also for *in vivo* recording. As far as synaptic plasticity goes and LTP similar to that in the hippocampus of vertebrates is present in the vertical lobes (89).

Also arthropods are able to perform complex learning (social learning). In insects the area responsible for learning and memory are the mushroom bodies, a plastic area that in the fruit fly is also involved in the regulation of sleep (70). Crayfish brain also shows a dynamic, stimulus-dependent plasticity in the visual interneurons of the brain, that form a complex distributed network (90). Thus, from the analyses of the life styles and brain complexity of these invertebrates, it's possible to state that many of them have in common plastic areas in the brain, and the capacity to carry out complex learning (91,92,93,94,95,96) the only exception being the jellyfish that can perform only simple learning such as habituation (97).

My conclusion is that, whatever the basal function of sleep is, such a role should emerge from a rigorous phylogenetic analysis. Thus in the future a broad

Sleep and its possible role in learning

phylogeny offers the possibility of testing the various ideas about the relationship between sleep and its function (e.g. memory trace processing, learning, synaptic-weight regulation, metabolism) and the coupling between plasticity and learning. A work of some importance in the future would be to establish if there are different kinds or 'flavours' of sleep in different animals and if so whether they are really homologous entities (i.e. have emerged from the same origins) or alternatively if 'it' represents several convergent paralogous processes. This question can only be approached by an accurate description of the behavioural and neurophysiological phenomena coupled with a good knowledge of the genes involved.

12. ACKNOWLEDGMENTS

S. Piscopo was supported by Marine Genomics Europe. The author would like to thank Dr. Gianna Nardi and Dr. Wiebe Kooistra for their kind help in reading and translating German language articles, Dr Carlo Di Cristo and Mr. Gabriele Ferrandino for their help with figure 1, Dr Antonio Piscopo for helpful comments, and Dr Euan R Brown for support and discussion.

13. REFERENCES

1. Campbell, S.S., Tobler, I.: Animal sleep: a review of sleep duration across phylogeny. *Neurosci Biobehav Rev.* 8,269-300 (1984)
2. Lesku, J.A., Rattenborg, N.C., Amlaner, C.J.: The evolution of sleep: a phylogenetic approach. *Sleep: A Comprehensive handbook*, ed. By T.Lee-Chiong 49-61 (2006)
3. Berger, H.: Uber das electroencephalogramm des Menschen. *J. Psychol. Neurol.* 40,160-179 (1930)
4. Aserinsky, E. and Kleitman, N.: Regular occurring periods of eye motility and concomitant phenomena during sleep. *Science* 118, 273-2 (1953)
5. Rojas-Ramírez, J.A., Tauber, E.S.: Paradoxical sleep in two species of avian predator (Falconiformes). *Science.* 167,1754-5 (1970)
6. Jouvet, M.: *La natura del sogno* (Ed. Theoria) Roma, Italy (1991)
7. Siegel, J.M.: Phylogeny and the function of REM sleep. *Behav. Brain. Res.* 69,29-34 *Proc Natl Acad Sci U S A* (1995)
8. Nicol, S.C., Andersen, N.A., Phillips, N.H., Berger, R.J.: The echidna manifests typical characteristics of rapid eye movement sleep. *Neurosci. Lett.* 283,49-52. (2000)
9. Siegel, J.M., Manger, P.R., Nienhuis, R., Fahringer, H.M., Shalita, T., Pettigrew, J.D.: Sleep in the platypus. *Neuroscience* 91,391-400 (1999)
10. Mukhametov, L.M., Polyakova, I.G.: EEG investigation of sleep in porpoises (*Phocoena phocoena*). *J High Nerve Activ* 31,333-339 (1981)
11. Tauber, E.S., Roffwarg, H.P., Weitzman, E.D.: Eye movements and electroencephalogram activity during sleep in diurnal lizard. *Nature* 212,1612-1613 (1966)
12. Melesio-Nolasco, S., Ayala Guerrero, F.: The phylogeny of sleep: fishes and amphibians. *Bol Estud Med Biol.* 32,101-7 (1983)
13. Garm, A., O'Connor, M., Parkefelt, L. and Nilsson, D.E.: Visually guided obstacle avoidance in the box jellyfish *Tripedalia cystophora* and *Chiropsella bronzie*. *Journ of Exp Biol.* 210,3616-3623 (2007)
14. Raizen, D.M., Zimmerman, J.E., Maycock, M.H., Ta, U.D., You, Y.J., Sundaram, M.V., Pack, A.I.: Lethargus is a *Caenorhabditis elegans* sleep-like state. *Nature.* 451,569-72 (2008)
15. Young, J.Z.: The distributed tactile memory system of Octopus. *Proceedings of the Royal Society of London, Series B*, 218, 135-176 (1983)
16. Young, J.Z.: Computation in the learning system of cephalopods. *Biological Bulletin* 180, 200-208 (1991)
17. Brown, E.R., Piscopo, S., De Stefano, R., Giuditta, A.: Brain and behavioural evidence for rest-activity cycles in *Octopus vulgaris*. *Behav Brain Res.* 172,355-9 (2006)
18. Tobler, I.: Effect of forced locomotion on the rest-activity cycle of the cockroach. *Behav Brain Res.* 8,351-60 (1983)
19. Tobler, I. and Stalder, J.: Rest in the scorpion - A sleep-like state? *J. Comp. Physiol.* 163, 227-235 (1988)
20. Tobler, I., Neuner-Jehle, M.: 24-h variation of vigilance in the cockroach *Blaberus giganteus*. *J Sleep Res.* 1,231-239 (1992)
21. Hendricks, J.C., Finn, S.M., Panckeri, K.A., Chavkin, J., Williams, J.A., Sehgal, A., Pack, A.: Rest in *Drosophila* is a sleep-like state. *Neuron.* 25,129-38 (2000)
22. Adam, K. and Oswald, I. : Sleep is for tissue restoration. *J. R. Coll. Physic.* 11, 376-388 (1977)
23. Takahashi, Y., Kipnis, D.M. and Doughaday, W.H.: Growth hormone secretion during sleep. *F. Clin. Invest.* 47, 2079-2090 (1968)
24. Brodskii, V.Y., Gusatinskii, V.N., Kogan, A.B. and Nechaeva, N.V.: Variations in the intensity of H³-leucine incorporation into proteins during slow-wave and paradoxical phases of natural sleep in the cat associative cortex. *Dokl. Akad. Nauk SSSR*, 748-750 (1974)

Sleep and its possible role in learning

25. Walker, J.M. and Berger, R.J.: Sleep as an adaptation for energy conservation functionally related to hibernation and shallow torpor. *Prog. Brain Res.* 53,255-278 (1980)
26. Berger, R.J. and Phillips, N.H.: Energy conservation and sleep. *Behav Brain Res* 69,65-73 (1995)
27. McGinty, D. and Szymusiak, R.: Keeping cool: a hypothesis about the mechanisms and functions of slow-wave sleep. *Trends in Neurosci* 13,480-487 (1990)
28. Inoue, S., Honda, K. and Komoda, Y.: Sleep as neuronal detoxification and restitution. *Behav Brain Res* 69,91-96 (1995)
29. Chen, C. and Tonegawa, S.: Molecular genetic analysis of synaptic plasticity, activity-dependent neural development, learning, and memory in the mammalian brain. *Annual Rev of Neurosci.* 20,157-184 (1997)
30. Kolb, B. and Whishaw, I.Q.: Brain plasticity and behavior. *Annual Rev of Psych* 49,43-64 (1998)
31. Kavanau, J.L.: Memory, sleep and the evolution of mechanisms of synaptic efficacy maintenance. *Neurosci.* 79,7-44 (1997)
32. Ebbinghaus, H.: *Memory. A Contribution to Experimental Psychology.* Eds Teachers College, Columbia University New York: (1913)
33. Roffwarg, H.P., Muzio, J.N., Dement, W.C.: Ontogenetic Development of the Human Sleep-Dream Cycle. *Science.* 152,604-619 (1966)
34. Mirmiran, M. and Ariagno, R.L.: Role of REM sleep in brain development and plasticity in "Sleep and Brain plasticity" edited by Maquet P., Smith C. and Stickgold R Eds. Oxford University Press UK 181-188 (2003)
35. Frank, M.G. and Stryker, M.P.: The role of sleep in the development of central visual pathways. in "Sleep and Brain plasticity" edited by Maquet P., Smith C. and Stickgold R. Eds Oxford University Press UK 189-208 (2003)
36. Jenkis, J.G. and Dallenbach, K.M.: Obliviscence during sleep and waking. *Am. J. Psychol.* 35,605-612 (1924)
37. Maquet, P.: The role of sleep in Learning and memory. *Science* 294,1048-1052 (2001)
38. Bloch, V.: Facts and hypothesis concerning memory consolidation processes. *Brain Research* 24,561-575 (1970)
39. Bloch, V.: L'activation cérébrale et la fixation mnésique. *Arch. Ital. Biol.* 3,577-590 (1973)
40. Bloch, V. and Fishbein, W.: Sleep and psychological function: memory. in G. C. Lairy and P. Salzarulo. Experimental study of human sleep: methodological problems., Eds Elsevier Scientific, Amsterdam: Netherlands: 157-173 (1975)
41. Crick, F. and Mitchison, G.: The function of dream sleep. *Nature* 304,111-114 (1983)
42. Maquet, P., Laureys, S., Peigneux, P., Fuchs, S., Petiau, C., Phillips, C., Aerts, J., Del Fiore, G., Degueldre, C., Meulemans, T., Luxen, A., Franck, G., Van Der Linden, M., Smith, C., Cleeremans, A.: Experience-dependent changes in cerebral activation during human REM sleep. *Nature Neuroscience* 3,831-836 (2000)
43. Stickgold, R., James, L., Hobson, J.A.: Visual discrimination learning requires sleep after training. *Nature Neuroscience* 3,1237-1238 (2000)
44. Vertes, R.P., Eastman, K.E.: The case against memory consolidation in REM sleep. *Behav. Brain Sci.* 23,867 (2000)
45. Siegel, J.M.: The REM Sleep-Memory Consolidation Hypothesis. *Science* 294, 1058-1063 (2001)
46. Stickgold, R.: Human studies of sleep and off-line memory reprocessing in "Sleep and Brain plasticity" edited by Maquet P., Smith C. and Stickgold R. Eds Oxford University Press UK 41-64 (2003)
47. Peigneux, P., Laureys, S., Cleeremans, A. and Maquet, P.: Cerebral correlates of memory consolidation during human sleep: contribution of functional neuroimaging in "Sleep and Brain plasticity" edited by Maquet P., Smith C. and Stickgold R. Eds Oxford University Press UK 209-224 (2003)
48. Benington, J.H., Frank, M.G.: Cellular and molecular connections between sleep and synaptic plasticity. *Prog in Neurobiol* 69,71-101 (2003)
49. Seymour, J.E., Carrette, T.J., Sutherland, P.A.: Do box jellyfish sleep at night? *Med J* 181,707 (2004)
50. Kavanau, J.L.: Is sleep's 'supreme mystery' unrevealing? An evolutionary analysis of sleep encounters no mystery; nor does life's earliest sleep, recently discovered in jellyfish. *Med Hypotheses.* 66,3-9 (2006)
51. Packard, A.: Cephalopods and fish: the limits of convergence. *Biological Reviews* 47, 241-307 (1972)
52. Fiorito, G., Scotto, P.: Observational learning in Octopus vulgaris. *Science* 256, 545-547 (1992)
53. Altman, J.S.: Underwater Association Report. Eds J.N. Lythgoe and J.D. Woods UK 77-83 (1966-1967)
54. Kayes, R.J.: The daily activity pattern of Octopus vulgaris in a Natural Habitat. *Mar. Behav. Physiol.* 2,337-343 (1974)
55. Cobb, C.S., Pope, S.K. and Williamson, R.: Circadian rhythms to light-dark cycles in the lesser octopus, *eledone cirrhosa*. *Mar. Fresh. Behav. Physiol.* 26,47-57 (1995)

Sleep and its possible role in learning

56. Bradley, E.A. and Young, J.Z.: Are there Circadian Rhythms in Learning by Octopus?. *Behav Biology* 13,527-531 (1975)
57. Lindauer, M.: Ein Beitrag zur Frage der Arbeitsteilung im Bienenstaat. *Z Vergl Physiol* 34,299-345 (1952)
58. Kaiser, W., Steiner-Kaiser, J.: Neuronal correlates of sleep, wakefulness and arousal in a diurnal insect. *Nature*. 24,707-9 (1983)
59. Sauer, S., Kinkelin, M., Herrmann, E., Kaiser, W.: The dynamics of sleep-like behaviour in honey bees. *J Comp Physiol A Neuroethol Sens Neural Behav Physiol*. 189,599-607 (2003)
60. Sauer, S., Herrmann, E., Kaiser, W.: Sleep deprivation in honey bees. *J Sleep Res*. 13,145-52 (2004)
61. Stephenson, R., Chu, K.M. and Lee, J.: Prolonged deprivation of sleep-like rest raises metabolic rate in the Pacific beetle cockroach, *Diploptera punctata* (Eschscholtz). *J. Exp. Biol*. 210,2540-2547 (2007)
62. Rechtschaffen, A., Bergmann, B.M.: Sleep deprivation in the rat: an update of the 1989 paper. *Sleep*. 25,18-24 (2002)
63. Ramón, F., Hernández-Falcón, J., Nguyen, B., Bullock, T.H.: Slow wave sleep in crayfish. *Proc Natl Acad Sci U S A*. 101,11857-61 (2004)
64. Mendoza-Angeles, K., Cabrera, A., Hernández-Falcón, J., Ramón, F.: Slow waves during sleep in crayfish: a time-frequency analysis. *J Neurosci Methods*. 162,264-71 (2007)
65. Shaw, P.J., Cirelli, C., Greenspan, R.J., Tononi, G.: Correlates of sleep and waking in *Drosophila melanogaster*. *Science*. 287,1834-7 (2000)
66. Seugnet, L., Boero, J., Gottschalk, L., Duntley, S.P., Shaw, P.J.: Identification of a biomarker for sleep drive in flies and humans. *Proc Natl Acad Sci U S A*. 103,19913-8 (2006)
67. Zimmerman, J.E., Mackiewicz, M., Galante, R.J., Zhang, L., Cater, J., Zoh, C., Rizzo, W., Pack, A.I.: Glycogen in the brain of *Drosophila melanogaster*: diurnal rhythm and the effect of rest deprivation. *J Neurochem*. 88,32-40 (2004)
68. Zimmerman, J.E., Rizzo, W., Shockley, K.R., Raizen, D.M., Naidoo, N., Mackiewicz, M., Churchill, G.A., Pack, A.I.: Multiple mechanisms limit the duration of wakefulness in *Drosophila* brain. *Physiol Genomics*. 27,337-50 (2006)
69. Nitz, D.A., van Swinderen, B., Tononi, G., Greenspan, R.J.: Electrophysiological correlates of rest and activity in *Drosophila melanogaster*. *Curr Biol*. 12,1934-40. (2002)
70. Pitman, J.L., McGill, J.J., Keegan, K.P., Allada, R.: A dynamic role for the mushroom bodies in promoting sleep in *Drosophila*. *Nature* 441,753-6 (2006)
71. Cirelli, C.: Searching for sleep mutants of *Drosophila melanogaster*. *Bioessays*. 25,940-9 (2003)
72. Huber, R., Hill, S.L., Holladay, C., Biesiadecki, M., Tononi, G., Cirelli, C.: Sleep homeostasis in *Drosophila melanogaster*. *Sleep* 27,628-39 (2004)
73. Cirelli, C., Bushey, D., Hill, S., Huber, R., Kreber, R., Ganetzky, B., Tononi, G.: Reduced sleep in *Drosophila* Shaker mutants. *Nature*. 434,1087-92 (2005)
74. Cirelli, C., La Vaute, T.M., Tononi, G.: Sleep and wakefulness modulate gene expression in *Drosophila*. *J Neurochem*. 94,1411-9 (2005)
75. Andretic, R., van Swinderen, B., Greenspan, R.J.: Dopaminergic modulation of arousal in *Drosophila*. *Curr Biol*. 15,1165-75 (2005)
76. van Swinderen, B., Andretic, R.: Arousal in *Drosophila*. *Behav Processes*. 64,133-144 (2003)
77. Kume, K., Kume, S., Park, S.K., Hirsh, J., Jackson, F.R.: Dopamine is a regulator of arousal in the fruit fly. *J Neurosci*. 25,7377-84 (2005)
78. Bushey, D., Huber, R., Tononi, G., Cirelli, C.: *Drosophila* Hyperkinetic mutants have reduced sleep and impaired memory. *J Neurosci*. 27,5384-93 (2007)
79. Cirelli, C., Bushey, D.: Sleep and wakefulness in *Drosophila melanogaster*. *Ann N Y Acad Sci*.;1129:323-9 (2008)
80. Ganguly-Fitzgerald, I., Donlea, J., Shaw, P.J.: Waking experience affects sleep need in *Drosophila*. *Science*. 313,1775-81 (2006)
81. Hendricks, J.C.: Invited review: Sleeping flies don't lie: the use of *Drosophila melanogaster* to study sleep and circadian rhythms. *J Appl Physiol*. (2003) Apr;94(4):1660-72;
82. Mackiewicz M, Naidoo N, Zimmerman JE, Pack AI.: Molecular mechanisms of sleep and wakefulness. *Ann N Y Acad Sci*.;1129:335-49 (2008)
83. Xu, X. and Norell, M.A.: A new troodontid dinosaur from China with avian-like sleeping posture. *Nature* 431,838-841 (2004)
84. Nilsson, D.E., Gislén, L., Coates, M.M., Skogh, C., Garm, A.: Advanced optics in a jellyfish eye. *Nature* 435,201-5 (2005)
85. Yatsu, N.: Notes on the physiology of *Charybdea rastonii*. *J.Coll.Sci. Imp. Univ. Tokyo* 40,1-12 (1917)

Sleep and its possible role in learning

86. Lewis, C. and Long, T.A.F.: Courtship and reproduction in *Carybdea sivickisi* (Cnidaria Cubozoa). *Mar. Biol.* 147,477-483 (2005)
87. Frank, M.G. and Heller, H.C.: Development of REM sleep and slow wave sleep in the rat. *American J of Physiol.* 272, R1792-R1799 (1997)
88. Mori, I., Sasakura, H. and Kuhara, A.: Worm thermotaxis: a model system for analyzing thermosensation and neural plasticity. *Curr Opin Neurobiol* 17,1-8 (2008)
89. Hochner, B., Brown, E.R., Langella, M., Shomrat, T., Fiorito, G.: A learning and memory area in the octopus brain manifests a vertebrate-like long-term potentiation. *J Neurophysiol.* 90,3547-54 (2003)
90. Wood, H.L., Glantz, R.M.: Distributed processing by visual interneurons of crayfish brain. II. Network organization and stimulus modulation of synaptic efficacy. *J Neurophysiol* 43,741-53 (1980)
91. Tempel, B.L., Bonini, N., Dawson, D.R., Quinn, W.G.: Reward learning in normal and mutant *Drosophila*. *Proc Natl Acad Sci U S A* 80,1482-6 (1983)
92. Ye, H.Y., Ye, B.P., Wang, D.Y.: Learning and learning choice in the nematode *Caenorhabditis elegans*. *Neurosci. Bull* 22,355-360 (2006)
93. Watanabe, H., Mizunami, M.: Classical conditioning of activities of salivary neurones in the cockroach. *J Exp Biol.* 209,766-79 (2006)
94. Kawai, N., Kono, R., Sugimoto, S.: Avoidance learning in the crayfish (*Procambarus clarkii*) depends on the predatory imminence of the unconditioned stimulus: a behavior systems approach to learning in invertebrates. *Behav Brain Res* 150,229-37 (2004)
95. Kim, Y.C., Lee, H.G., Han, K.A.: Classical reward conditioning in *Drosophila melanogaster*. *Genes Brain Behav.* 6,201-7 (2007)
96. Boal, J.G., Dunham, A.W., Williams, K.T., Hanlon, R.T.: Experimental evidence for spatial learning on octopuses (*Octopus bimaculoides*). *J Comp Psychol.* 114,246-52 (2000)
97. Johnson, M.C., Wuensch, K.L.: An investigation of habituation in the jellyfish *Aurelia aurita*. *Behav Neural Biol.* 61,54-9 (1994)

Key Words: Sleep, Phylogeny, Invertebrate, Cnidarians, Arthropods, Molluscs, Cephalopods, Nematodes, Review

Send correspondence to: Stefania Piscopo, Laboratorio di Fisiologia Animale ed Evoluzione, Stazione Zoologica "A.Dohrn", Napoli, Italy, Tel: 39- 081-5833228, Fax: 081-764-1355, E-mail: piscopo@szn.it

<http://www.bioscience.org/current/vol1S.htm>