OPINION

# Neurogenesis in the adult brain: death of a dogma

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For over 100 years a central assumption in the field of neuroscience has been that new neurons are not added to the adult mammalian brain. This perspective examines the origins of this dogma, its perseverance in the face of contradictory evidence, and its final collapse. The acceptance of adult neurogenesis may be part of a contemporary paradigm shift in our view of the plasticity and stability of the adult brain.

Until very recently, a central dogma of neuroscience has been that new neurons are not added to the adult mammalian brain. For more than 100 years it has been assumed that neurogenesis, or the production of new neurons, occurs only during development and stops before puberty[1–3]. Indeed, there are few views of the brain that have persisted for so long with so little successful challenge.

This perspective examines the origins of this dogma and discusses how it has persisted even in the face of new techniques that were able to disprove it, how it finally narrowed and is now in disarray. This decline in belief in the stability of the neuronal population seems to be part of a more general paradigm shift[4] that recognizes the plasticity of the adult brain and its structural modulation by experience.

## The dogma up to mid-century

By the end of the nineteenth century, the idea that the brain of the adult mammal remains structurally constant was already universally held by the main figures of the time, including Koelliker[5], His[6] and Cajal[1,7]. What were the origins of this view? Koelliker, His and others had described in detail the development of the central nervous system of humans and other mammals. They found that the structure of the brain remained fixed from soon after birth. Because the elaborate architecture of the brain remained constant in appearance, the idea that neurons were continually added to it was, understandably, inconceivable. Similarly, Ramón y Cajal and others had also described the different phases in the development of the neuron, terminating with the multipolar structure characteristic of the adult. As neither mitotic figures nor these developmental stages had been seen in the adult brain, the possibility of continuing neuronal addition to the adult brain was rarely, if ever, seriously entertained.

In the first half of the twentieth century, there were occasional reports of postnatal neurogenesis in mammals. For example, Schaper[8,9] claimed there were "indifferent" cells that were widely distributed in the brain from teleost to human, even into adult life, and that these indifferent cells could become either neurons or glia; and Levi[10] reported mitosis in small, but not large, neurons in brain-injured guinea pigs. At about the same time, Hamilton[11] saw mitosis in four-day-old rats; and Allen[12] found mitotic figures in the cerebrum of the rat until at least 120 days after birth. Moreover, Sugita[13] counted an increased number of cortical neurons in the rat over the first 20 postnatal days; and Bryans[14], using colchicine to freeze mitosis, detected cell division in the brains of rats that were at least one year old. In several of these studies, mitotic figures were found lining the walls of the lateral ventricle, in the subependymal layer (now termed the subventricular zone) of adult rats. Consequently, the possibility that the new cells arising in the subependymal layer might migrate into the cerebrum to form mature neurons was raised[11,13,14].

However, in these and similar studies, it was unclear whether the cells undergoing mitosis subsequently became glia or neurons. As Ramón y Cajal[1] put it in a critique of the earlier of these studies:

"Unfortunately, however, none of the methods used by these investigators are capable of distinguishing absolutely a multiplying neuroglia cell from a small mitotic neuron."

These scattered reports, raising the possibility of adult mammalian neurogenesis, tended to be ignored by textbooks and were rarely cited. Presumably this was because of the weight of authority opposed to the idea and the inadequacy of the available methods both for detecting cell division and for distinguishing glia from small neurons.

## Tritiated thymidine

An important advance in the study of neurogenesis came in the late 1950s with the introduction of [$^3$H]-thymidine autoradiography. [$^3$H]-thymidine is incorporated into the DNA of dividing cells. Therefore, the progeny of cells that had just divided could be labelled, and their time and place of birth determined (FIG.1a). Initially, this new method was applied almost exclusively to the study of the developing rodent, particularly by Richard Sidman and his students[15]. Their emphasis on using this method to study pre- and perinatal development, rather than looking across the life span of the animal, reflected the persistence of the belief that neurogenesis did not occur in the adult mammal.

In 1961, [$^3$H]-thymidine autoradiography was used for the first time to study proliferation in the adult brain by Smart[16]. Whereas he found cells that arose from the subependymal layer and migrated into the adjacent brain to become neurons and glia in three-day-old

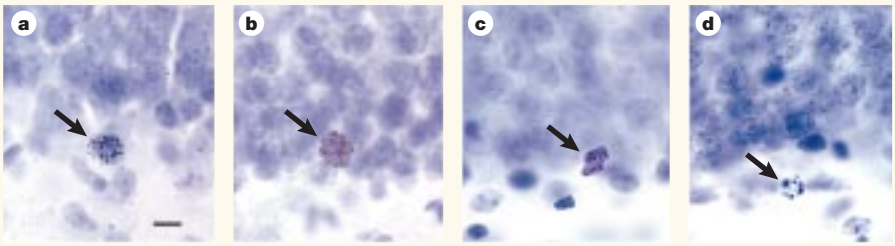


Figure 1 | **Cell birth and death in adulthood.** Light photomicrographs from granule cell layer of dentate gyrus stained with cresyl violet. **a** | Rat [$^3$H]-thymidine-labelled cell (arrow). **b** | Rat BrdU-labelled cell (arrow). **c** | Macaque BrdU-labelled mitotic figure in anaphase (arrow). **d** | Macaque pyknotic (dying) cell (arrow). Scale bar in **a** is 5μm and applies to **a–d**. (Image courtesy of N. Hastings and E. Gould.)

mice, he was unable to obtain clear evidence for a similar process in adults.

Starting in the early 1960s, Joseph Altman began publishing a series of papers[17–23] in which he reported thymidine autoradiographic evidence for new neurons in various structures in the young and adult rat, including the neocortex[18,21], dentate gyrus[18,19] and olfactory bulb[23]. He also reported new neurons in the neocortex and elsewhere in the adult cat[18]. He argued that the new neurons were "microneurons" — granule or stellate cells with short axons — and suggested that they were crucial in learning and memory[22]. Although published in the most prestigious journals of the time, such as the *Journal of Comparative Neurology*, *Science* and *Nature*, these findings were ignored or dismissed as unimportant for over two decades. A typical contemporary treatment of Altman's reports of adult neurogenesis was Paul Weiss's[24] remark: "sporadic residual straggler neurons have been reported" followed by a reference to a comprehensive paper by Altman[22].

The neglect of Altman's demonstration of adult neurogenesis is a classic case of a discovery made 'before its time'. There seem to have been several reasons why Altman's work was ignored. First, the available techniques were not really adequate for an unambiguous demonstration that the adult-generated cells were neurons rather than glia. The small nuclei of the microneurons could not be easily distinguished from glial nuclei and a grazed [$^3$H]-thymidine-labelled neuroglia cell could have caused an underlying neuron to appear labelled. Second, the results implied a considerable and therefore unlikely migration from the ventricle to, for example, the olfactory bulb or the cerebral cortex. Last, an important reason for the neglect of Altman's work may have been that he was a self-taught postdoctoral fellow working on his own in a Psychology Department (at Massachusetts Institute of Technology (MIT)) and that his work was attempting to overturn a central and, by then, universally held tenet of neuroscience. Altman was not granted tenure at MIT and moved to Purdue University where he eventually turned to more conventional developmental questions[25], possibly because of the lack of recognition of his work on adult neurogenesis. As late as 1970, an authoritative textbook of developmental neuroscience[3] stated that "... there is no convincing evidence of neuron production in the brains of adult mammals".

## Electron microscopy

Fifteen years after Altman's first report, direct support for his claim of adult neurogenesis came from a series of electron microscopy studies by Michael Kaplan and his co-authors. First, they showed that [$^3$H]-thymidine labelled cells in the dentate gyrus and olfactory bulb of adult rats have the ultrastructural characteristics of neurons, such as dendrites and synapses, but not of astrocytes or oligodendrocytes[26,27]. Then Kaplan[28,29] reported autoradiographic and ultrastructural evidence for a few new neurons in the cerebral cortex of adult rats, again confirming the earlier claims of Altman[18,21]. Finally, he showed mitosis in the subventricular zone of adult macaque monkeys by again combining [$^3$H]-thymidine labelling and electron microscopy[30]. During this research period, Kaplan was, successively, a graduate student at Boston University, a post-doctoral fellow at Florida State University and an assistant professor at the University of New Mexico. He then left the field, becoming a medical student (M.K., personal communication). In spite of his evidence for adult neurogenesis, Kaplan's work had little effect at the time, as measured by citations or follow-up studies. Again, as in Altman's case, publication in prestigious and rigorously reviewed journals, such as *Science*, the *Journal of Comparative Neurology* and the *Journal of Neuroscience*, by an unknown figure was not sufficient to make any marked dent in the dogma. Similiar cases are well documented in the history of science[31].

## Lack of evidence

Another reason for the small impact of Kaplan's work was probably a study presented at a meeting in 1984 and published the following year[2,32]. Pasko Rakic, the author of the study, was (and still is) Professor at Yale Medical School and arguably the leading student of primate brain development. He carried out a [$^3$H]-thymidine study of adult rhesus monkeys in which he examined "all major structures and subdivisions of the brain including the visual, motor, and association neocortex, hippocampus, olfactory bulb". Rakic found "not a single heavily labelled cell with the morphological characteristics of a neuron in any brain of any adult animal" and concluded that "all neurons of the rhesus monkey brain are generated during prenatal and early postnatal life"[2,32].

Rakic's papers[2,32] had a profound influence on the development of the field. Subsequent work in adult rhesus monkeys by Eckenhoff and Rakic[33], using a combination of thymidine autoradiography, electron microscopy and an immunocytochemical marker for astroglia (but not for neurons), also found no adult-generated neurons in the primate dentate gyrus. Furthermore, they questioned the reports of adult neurogenesis in rats with the suggestion that rats never stop growing and so never become adults. (In fact, there are strains of rats that do stop growing and also show adult neurogenesis[34,35] but this was not known at the time.) For Eckenhoff and Rakic, the supposed lack of adult neurogenesis in primates made sense, because, "a stable population of neurons may be a biological necessity in an organism whose survival relies on learned behaviour acquired over a long period of time". Furthermore, Rakic suggested that the "social and cognitive behaviour" of primates may require the absence of adult neurogenesis[2]. As discussed elsewhere, humans show a basic need to distinguish themselves from other animals[36,37]. Although neuroscientists have often tried to make this distinction in terms of brain structure or function[37], this may be the only time that the social and cognitive differences between primates and non-primates was attributed to the presence or absence of adult neurogenesis and, more generally, to the structural stability of the brain. Moreover, as described below, by 1999, when new techniques were available, Rakic himself published evidence for neurogenesis in the dentate gyrus of adult primates[38].

The following sections of this paper discuss three developments that eventually led to the general acceptance that adult-generated neurons are added to one mammalian brain region, namely the hippocampus of adult

rodents, and that this was probably an interesting and important phenomenon. The first development was a series of experiments that unambiguously showed neurogenesis in adult birds, which were carried out under the leadership of Fernando Nottebohm, already a highly respected scientist. The second development was the introduction of new methods for labelling new cells and for distinguishing neurons from glia. Finally, demonstrations that neurogenesis could be up- and downregulated by important psychological variables such as stress, environmental complexity and learning as well as by hormones, raised the possibility that adult hippocampal neurogenesis was more than some ontogenetic or phylogenetic vestige and might be important for cognition in higher animals. In the final sections of the paper I return to the questions of adult primate neurogenesis and the possible functions of adult-generated neurons.

### Avian neurogenesis

Starting in the late 1960s, Nottebohm and his colleagues at Rockefeller University began a systematic analysis of the neural basis of song learning in birds. They discovered a set of brain mechanisms that are crucial for bird song and showed how the volume of two nuclei were a function of variables such as sex, sexual maturity, song complexity, species, testosterone level and season[39–41]. The seasonal and hormonal changes in the volume of these song-related nuclei were so great in some species that Nottebohm set out to examine the possibility that these changes were due to fluctuations in the actual number of neurons in the adult avian brain.

In a series of elegant experiments, starting in the early 1980s, Nottebohm and his colleagues showed that , indeed, thousands of new neurons are added every day to the avian brain. They did so by, first, showing the production of new cells with thymidine labelling[42]; second, producing ultrastructural evidence that the new cells were neurons receiving synapses[43]; and last, in a technical tour de force, showing that the putative neurons responded to sound with action potentials[44]. In subsequent studies, they showed that the axons of new neurons extended over long distances, that neuronal birth and death proceeded in parallel, that in both singing and non-singing species neurogenesis was widespread throughout the avian forebrain — including the hippocampus — and that in the latter structure it was modulated by environmental complexity and learning experience[39–47].

In spite of this unassailable evidence of neurogenesis in parts of the adult bird brain known to be homologous to primate cerebral cortex and primate hippocampus, these studies tended to be viewed as irrelevant to the primate or even to the mammalian brain. Rather, the evidence for avian neurogenesis was viewed as an exotic specialization related to the necessity for flying creatures to have light cerebrums and to their seasonal cycles of singing.

> "…None of the methods used by these investigators are capable of distinguishing absolutely a multiplying neuroglia cell from a small mitotic neuron." (Cajal, 1913)

### New techniques

Beginning around the 1990s, there were several developments that finally established the reality of neurogenesis in the dentate gyrus of the adult rat. One was the demonstration that the new cells in the rat dentate gyrus extend axons into the mossy fibre pathway[48].

Another important development was the introduction of the synthetic thymidine analogue BrdU (5-bromo-3'-deoxyuridine). Like thymidine, BrdU is taken up by cells during the S-phase of mitosis and is a marker of proliferating cells and their progeny. BrdU labelling can be visualized with immunocytochemical techniques (FIG. 1b) and does not require autoradiography[49]. This technique allows stereological estimation of the total number of cells as well as demonstration that the new cells express markers of specific cell types. Gage and his colleagues were the first to use BrdU labelling[35] and stereology[50] in the study of adult neurogenesis in the rodent.

Another important advance was the use of cell-type specific markers for the immunohistochemical identification of the newly generated cells. Among the markers for mature neurons are NSE (neuron specific enolase)[51,52], MAP-2 (microtubule-associated protein 2)[53,54], TuJ1 (class III beta-tubulin)[55] and NeuN (neuronal nuclei)[35,56]. Although some of these markers have been shown to stain non-neuronal cells under certain conditions (for example, NSE and MAP-2) and others do not stain all neuronal types (for example, NeuN and MAP-2)[56–59], the expression of several of these antigens in a population of adult-generated cells is considered good evidence that new neurons have been produced. Immature neurons can be marked with the Hu protein[60,61], PSA-NCAM (polysialylated-neural cell adhesion molecule)[62,63], TuJ1[63,64] and CRMP4 (collapsin response mediated protein 4), the antibody previously known as TOAD-64 (turned on after division 64)[65–67]. Some of these markers stain non-neuronal cells as well[68,69]. There are also markers for oligodendrocytes, such as CNP (2′,3′-cyclic nucleotide 3′ phosphodiesterase)[38,70] and O4 (oligodendrocyle cell surface marker no. 4)[38,71], and for astrocytes, such as GFAP (glial fibrillary acidic protein)[72,73] and S100β (a calcium binding protein)[74,75]. If a cell expresses neuronal markers and not glial markers, it constitutes further evidence that it is a neuron and not a glial cell. For examples of cell-type specific markers, see FIG. 2a–e.

It is interesting to note the apparent differences in the readiness to accept plasticity in different brain structures. A few years after Altman had reported postnatal neurogenesis in the rodent cerebral cortex[18] and dentate gyrus[18,19], he also claimed it for the olfactory bulb[23]. The strength (or weakness) of the evidence was about the same for the different areas. But the three claims had different fates. Adult neurogenesis in both the olfactory bulb[76] and dentate gyrus[26] was replicated early but, even by 1991, only the olfactory bulb result was included in Jacobson's authoritative text[77]. This may have reflected a greater willingness to accept plasticity in phylogenetically older and supposedly less 'cognitive' areas. The situation with the rodent cerebral cortex is still unclear. Kaplan[28,29] reported autoradiographic and ultrastructural evidence for cortical neurogenesis in the rat but recently Macklis and his colleagues did not find neurogenesis in the cortex of the mouse unless apoptosis was induced[61].

### Regulation of neurogenesis

The advent of new techniques meant that by the early 1990s, Altman's claims that new neurons were added to the adult dentate gyrus had been confirmed several times[26,72,78]. But was this phenomenon more than some ontogenetic lag or phylogenetic vestige? A series of studies soon revealed that dentate gyrus neurogenesis in the rodent could be modulated by experience and so might be important for cognitive function.

Gould and collaborators showed that acute and chronic stress decreased the number of adult generated neurons in the dentate gyrus. Adrenal steroids probably underlie this effect as stress increases adrenal steroid levels and glucocorticoids decrease the rate of neurogenesis[79]. Cameron and McKay extended these findings by showing that the decreased level of neurogenesis in the dentate gyrus during ageing[35] is due to an increase in glucocorticoid levels[80]. Chronic administration of

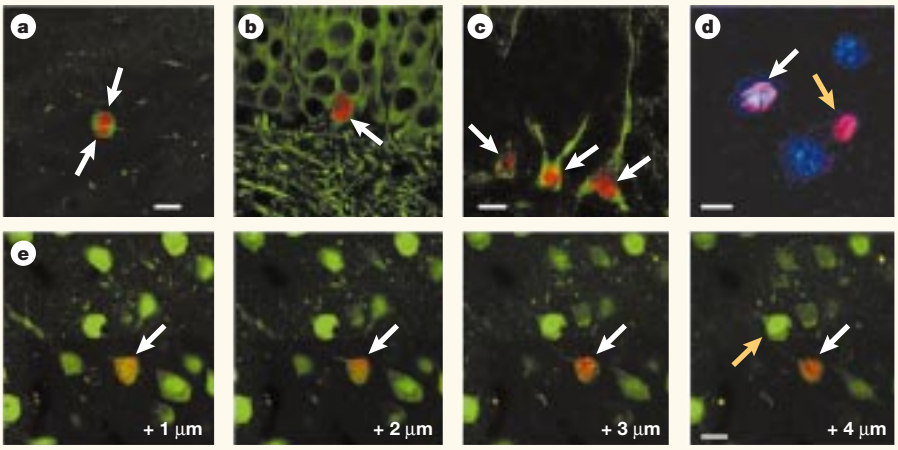


Figure 2 | **Cell-specific markers and neurogenesis.** Confocal laser scanning microscopic images of BrdU-labelled cells. **a** | Two cells (white arrows) double labelled for BrdU (red nuclear stain) and the oligodendrocyte marker CNP (green nuclear stain) from frontal cortex of adult macaque. The scale bar is 5 μm and also applies to b. **b** | BrdU-labelled cell (red nuclear stain, white arrow) co-labelled with the neuronal marker, microtubule-associated protein 2 (MAP-2) (green cytoplasmic stain) from the dentate gyrus of an adult macaque. **c** | Cells double labelled for BrdU (red nuclear stain, white arrows) and the neuronal marker, class III beta-tubulin (TuJ1) (green cytoplasmic stain) from the dentate gyrus of the adult rat. Scale bar is 5μm. **d** | Cell from posterior parietal cortex of adult macaque (white arrow) co-labelled with BrdU (red nuclear stain) and the retrograde tracer Fast Blue (blue cell-body marker). Yellow arrow points to BrdU-labelled cells not co-labelled with Fast Blue. The scale bar is 10 μm. **e** | Confocal laser scanning microscope images (z-series). 1 μm sections through a BrdU-labelled cell (white arrow) in prefrontal cortex of adult macaque showing co-localization throughout with the neuronal marker, neuronal nuclei (NeuN) (green nuclear and cytoplasmic stain). The yellow arrow indicates a cell labelled only with NeuN. The scale bar is 15 μm. (Image courtesy of E. Gould.)

morphine and heroin also diminishes neurogenesis but here the mechanism does not seem to involve corticosteroids[81].

On the other hand, there are several conditions that increase the number of adult-generated dentate gyrus cells. First, oestrogen stimulates the production of new immature neurons in the dentate gyrus whereas ovariectomy reduces it[66]. Second, environmental complexity increases the number of new hippocampal neurons as shown in birds in Nottebohm's laboratory[45], in mice in Gage's[50,82] and in rats in Eriksson's[83]. Curiously, just running in a wheel enhances the number of BrdU-labelled cells in the dentate gyrus[75,84], but whether this is due to enhanced environmental stimulation, reduction in stress or some peripheral effect of exercise, such as increased blood flow, is unclear[85].

Although 'enriched environments' presumably offer more opportunities for learning than do standard laboratory environments, there are many other differences between the two living conditions, such as the quantity and quality of social, visual, auditory, tactile and motor stimulation. To isolate the effects of learning, Gould *et al.* studied the influence of specific learning experiences on new neurons in the dentate gyrus of adult rats[86]. The number of new neurons was enhanced in animals that were trained on trace eye blink conditioning or spatial navigation learning, two hippocampal-dependent tasks. In contrast, no change in the number of new neurons was found in animals trained on tasks that are not hippocampus-dependent. Experience on hippocampus-dependent tasks extended survival rather than enhancing the production of new cells. However, the increased survival of new neurons may not occur unless the learning experience occurs at a specific time after cell production[75,87]. Furthermore, there is some suggestion that the enhancement may be confined to specific parts of the dentate gyrus[88].

In summary, there are several conditions that either increase or decrease adult neurogenesis in the dentate gyrus, indicating that adult neurogenesis may be important for hippocampal function.

## The dogma crumbles

The 1990s was a decade of increasing evidence for central nervous system plasticity. Examples include Greenough's work on the effect of environmental complexity on dendritic spines[89] and Kaas and Merzenich's demonstration of somatosensory receptive field reorganization after injury and experience[90,91]. Knudsen's analysis of the effect of early experience on the sensory organization of the owl tectum[92] and Weinberger's demonstration of the effect of auditory learning on auditory cortex[93] provided further support, as did McEwen's studies of the effects of hormones on the brain[94] and the continued growth of the long-term potentiation (LTP) field. These demonstrations of plasticity in the brains of adult mammals may have encouraged the renewed search for neurogenesis in the adult primate and made it a more plausible possibility than in the past.

At the end of the decade, adult neurogenesis was shown in the primate dentate gyrus. Using BrdU labelling, combined with neuronal markers, new neurons were reported in the dentate gyrus of the marmoset and the macaque by Gould *et al.*[95,96], in the macaque by Kornack and Rakic[38], and in humans by Ericksson *et al.*[52]. This latter study, which involved human cancer patients, was a turning point in the acceptance of adult neurogenesis as a real phenomenon, because it showed that even in the most complex and highly evolved brain, new neurons continue to be added throughout life. It is not yet clear if the rate or characteristics of neurogenesis varies within the primate order or between primates and rodents because comparable quantitative data are not yet available across the different species. Further evidence for adult neurogenesis in the hippocampus comes from studies in which progenitor cells capable of proliferation and neurogenesis have been isolated from the hippocampus, specifically in adult rats by Gage *et al.*[97] and in humans by Goldman and colleagues[54].

Recently, Gould and colleagues[98,99] reported that a relatively small number of new neurons were added to the neocortex of the adult macaque. In an extension of this work, the authors showed that most of these adult-generated cortical neurons have a transient existence[99]. For some methods used to show adult neuron birth and death, see FIGS 1,2.

So more than 100 years after the formulation of the neuron doctrine, it was finally clear that its corollary 'new neurons are not added to the adult mammalian brain' is false: even for adults, even for primates and, apparently, even for the cerebral cortex. The question now is what, if anything, are the functions of the new cells?

## A role in learning and memory?

Every day thousands of new neurons are added to the mammalian brain[100]. Although the new neurons are a minuscule proportion of the total population, their continual addition over a lifetime implies considerable structural change. The magnitude and ubiquity of adult neurogenesis across vertebrates suggests that it is functionally significant and not merely a vestige of development.

The available data indicate that the new cells may have lifetimes ranging from a few

days to the life of the animal[52,86]. There may be one population of adult-generated neurons that turns over with variable longevities and another, apparently much smaller, that is permanent. The turnover of most (or all) of the new neurons makes it unlikely that their function is to permanently replace dying cells that originated during development.

This section considers the possibility that adult generated cells have functions in learning and memory. In the next, more speculative section, some possible functions of new neurons in learning are discussed. Both sections focus on the hippocampus because a higher density of new neurons are generated there and more is known about them.

There are five considerations that, in my opinion, suggest that neurogenesis in adult mammals might be important for learning and memory. First, new neurons are added to structures crucial for learning and memory. These include, in many vertebrates, the hippocampus[100] (involved in spatial learning and consolidation of long term memory), and in macaques[98], lateral prefrontal cortex (involved in spatial short-term memory and visual-motor association), inferior temporal cortex (involved in visual learning) and posterior parietal cortex (involved in spatial learning) but not primary visual (striate) cortex, which is not usually considered to be an area involved in learning and memory.

Second, several conditions that decrease the proliferation of granule cells in the dentate gyrus, such as acute and chronic stress[79], increased levels of circulating corticosteroids[79], ageing[35,96] and opiate administration[81], also impair hippocampal-dependent learning such as spatial learning in a water maze[101]. So these learning impairments may be related to a diminished pool of adult-generated granule neurons.

Third, several conditions that increase the proliferation of hippocampal cells enhance learning on hippocampal-dependent tasks. These include increased oestrogen levels[66], increased environmental complexity[50,83] and wheel running[75,84]. The findings of van Praag *et al.*[84], that wheel running enhanced dentate gyrus LTP as well as dentate neurogenesis and spatial learning[84], are also consistent with the idea of an association between adult neurogenesis and learning. Finally, specific learning experience on hippocampal-dependent tasks extends the life of new cells in the dentate gyrus[86]. These results further support the possibility that adult neurogenesis in the hippocampus may be important for the learning functions of this structure. However, it should be noted that both the negative-modulators of neurogenesis, such as stress, and its positive-modulators, such as environmental complexity, have various effects other than those on neurogenesis, for example, changes in dendritic structure, in the synapse, in glia and in vasculature, any of which could contribute to the changes in learning performance[85,89,94,102].

Fourth, new neurons (or at least previously unused ones) are predicted by and required for some computational theories of learning. Many contemporary programmable models of learning have found that it is not feasible to use a fixed network. So they have either postulated that the network 'adds units'[103–105] or that there are a large set of previously unused neurons that can be randomly connected with the network[106].

"We may be in the midst of a revolution in our conception of the plasticity of the adult mammalian brain."

Fifth, adult-generated neurons may be similar to embryonic and early postnatal neurons in that they possess some properties that may make them particularly suitable to function in learning and memory[100,101]. For example, during development new neurons extend axons, even during migration, and rapidly make new synapses[107]. Similarly, adult-generated dentate gyrus neurons extend axons from four to ten days after their generation, indicating that they may be making synapses long before they become fully mature[108]. Perhaps they are particularly plastic and make connections more readily during this early stage[100,101]. Furthermore, granule cells show LTP of greater duration in younger rats compared with older rats[109]. Finally, granule cells that are presumed to have been generated in adulthood have a lower threshold for LTP induction, produce greater short-term facilitation, and are "more plastic and less prone to GABA-mediated inhibition" than older cells[110].

## Function: some speculations

In this more speculative section, I discuss some possible functions of adult-generated neurons in learning and memory. These neurons have temporal properties that may be related to the formation of memories. The daily addition of neurons to the brain may account for the fact that long-term memories are time-tagged, a phenomenon suggested by aspects of memory loss in human patients[111]. For example, in conditions such as Alzheimer's disease, there is a progressive retrograde loss of more remote memories. Moreover, in the recovery from traumatic amnesia, the oldest memories return first. These temporal gradients of amnesia imply that younger memories are different from older ones, in the sense that older memories are more resistant to interference by trauma and disease. The continual addition of new neurons suggests a possible mechanism for this phenomenon. There may be sets of cells that store a particular memory, with new adult-generated neurons continually added to such memory circuits. Therefore, the older the circuit for a particular memory, the more neurons it would have and the more resistant it might be to trauma because of greater redundancy, greater spatial dispersion or both. Such a scheme may account for the greater resistance of older memories to loss of (or interference with) retrieval. This hypothesis requires that there is a population of adult-generated neurons that has a lengthy survival time.

However, many adult-generated neurons seem to have a time-limited existence. This might be related to transient processes thought to be involved in memory[111]. One such process is the transformation of short-term memory into long-term memory. Perhaps the pattern of activity in circuits involving newly generated neurons represents the temporary storage of an event. If this pattern of activity persists, it may produce a lasting change in the older and permanent neurons that represent long-term memories, perhaps in their gene expression, resulting in a change in synaptic efficacy. After this 'consolidation' occurs, the adult-generated neurons may die, making room for new naive neurons that would then function similarly in consolidating new memories.

A specific role for the transient adult-generated neurons may possibly be in the time-limited storage functions attributed to the hippocampus. This structure seems to be essential for storage of declarative memories, although only for a limited time[111,112]. The function of the hippocampus in maintaining this information diminishes for older memories, which are thought to be established in the neocortical association areas. These temporary memory functions of the hippocampus could involve transient adult-generated hippocampal neurons. When these memories become permanently stored in neocortical circuits, the now unnecessary adult-generated neurons may die. In rats, many adult-generated hippocampal cells die within three weeks of their generation[72,86]. In macaques, many of the adult-generated neurons die within nine weeks[99]. These lifetimes correspond approximately to the estimated duration of hippocampal storage in the two species[111], a rough correlation which lends some plausibility to

the idea that transient hippocampal neurons may be important for the transient storage functions of the hippocampus.

## Concluding thoughts

The idea that new neurons are not added to the brains of adult mammals dates back to the neuron doctrine and the origins of modern neuroscience at the end of the nineteenth century. The tenacious persistence of this dogma in the face of empirical contradiction and its relatively recent demise illustrates, among other things, the strength of tradition and the difficulty that unknown and junior scientists have in challenging such traditions. It also suggests the necessity for new ideas to arise in a supportive matrix if they are to survive, and under scores the importance of new techniques. The general acceptance of adult neurogenesis today, at least in the dentate gyrus of the hippocampus, is suggestive of a paradigm shift. We may be in the midst of a revolution in our conception of the plasticity of the adult mammalian brain.

It should be stressed that the actual number of adult-generated neurons is a small proportion of the total population of neurons. But the existence of adult-generated neurons in the hippocampus (and probably elsewhere), and the possibility that these cells may function in learning and memory offer new mechanisms for information storage in the brain. It may be that learning and memory involve the development of entirely new circuits with new and previously unused elements as well as the modulation of older circuits and connections.

Finally, adult neurogenesis may also be relevant, in the long run, to the development of therapeutic strategies for the treatment of brain damage and disease.

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TIMELINE

# Transcranial magnetic stimulation and cognitive neuroscience

*Vincent Walsh and Alan Cowey*


Transcranial magnetic stimulation has been used to investigate almost all areas of cognitive neuroscience. This article discusses the most important (and least understood) considerations regarding the use of transcranial magnetic stimulation for cognitive neuroscience and outlines advances in the use of this technique for the replication and extension of findings from neuropsychology. We also take a more speculative look forward to the emerging development of strategies for combining transcranial magnetic stimulation with other brain imaging technologies and methods in the cognitive neurosciences.


Transcranial magnetic stimulation (TMS) is now an established investigative tool in the cognitive neurosciences[1–5], and several groups have begun to exploit its potential in the study of perception[6–16], attention[16,17], learning[18,19], plasticity[20–24], language[25–27] and awareness[28,29]. It is also finding applications in the study and treatment of movement disorders[30–32], epilepsy[33,34], depression[35–38], anxiety disorders[39–41], stuttering[42,43] and schizophrenia[44–47] (TIMELINE). Despite the breadth and depth of the published research, the considerations behind the use of TMS and its value in addressing neuropsychological questions remain poorly understood. In this article we confront some of the most common confusions about TMS and show how it can be used to complement and extend existing techniques. The use of TMS in clinical neurophysiological studies is highly advanced and has been reviewed elsewhere[48]. Likewise, parameters for the safe use of TMS have been established and have been documented extensively in other sources that are required reading for those contemplating the use of TMS[49–51]. Our aim is not to provide a technical introduction (which can be found in REFS 52–54). Here we focus on the role of TMS in the cognitive neurosciences and propose a conceptual framework for the future application of TMS to this area (FIG. 1).