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STATINS AND NEUROPROTECTION: A PRESCRIPTION TO MOVE THE FIELD FORWARD

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Abstract

There is growing interest in the use of statins, HMG-CoA reductase inhibitors, for treating specific neurodegenerative diseases (cerebrovascular disease, Parkinson's disease, Alzheimer's disease, multiple sclerosis) and possibly traumatic brain injury. Neither is there is a consensus on the efficacy of statins in treating the aforementioned diseases nor are the mechanisms of the purported statin-induced neuroprotection well-understood. Part of the support for statin-induced neuroprotection comes from studies using animal models and cell culture. Important information has resulted from that work but there continues to be a lack of progress on basic issues pertaining to statins and brain which impedes advancement in understanding how statins alter brain function. For example, there are scant data on the pharmacokinetics of lipophilic and hydrophilic statins in brain, statin-induced neuroprotection versus cell death and statins and brain isoprenoids. The purpose of this mini-review will be to examine those aforementioned issues and to identify directions of future research.

Keywords

Alzheimer's disease; Cerebrovascular disease; Cholesterol; HMG-CoA reductase; Isoprenoids; Neurodegeneration; Neuroprotection; Statins; Stroke

INTRODUCTION

Statins are universally recognized for their effectiveness in reducing serum cholesterol levels and reducing the morbidity and mortality associated with coronary heart disease 1,2. What has not received as much attention but is a topic of emerging interest is the efficacy of statins in the prevention or treatment of certain neurodegenerative diseases: e.g., cerebrovascular disease 3, Parkinson's disease 4, Alzheimer's disease 5, and multiple sclerosis 6. A recent report also indicates that statins may be effective in treating traumatic brain injury 7. There have been several recent and excellent reviews on statins and neuroprotection 5,8–12. Many of those reviews have covered clinical and experimental data both pro and con concerning the use of statins in neurodegenerative diseases. In this review, our intention is not to reexamine that body of data. Instead, the focus of this review will be on specific issues pertaining to statin use in animal and cell culture studies and recommendations to move the field forward. The issues which will be examined are: 1) the

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pharmacokinetics of lipophilic and hydrophilic statins in brain; 2) statin-induced neuroprotection versus cell death; and 3) statins and brain isoprenoids.

Pharmacokinetics of Lipophilic and Hydrophilic Statins in Brain

There have been no comprehensive studies published on the uptake, distribution and metabolism of statins in brain. A small number of studies have provided some data on statin levels in brain. One study of 12 human subjects found that lovastatin acid but not pravastatin was detected in the cerebrospinal fluid following a 5 day drug treatment and it was concluded that pravastatin did not pass the blood-brain barrier 13. The conclusion that pravastatin did not pass the blood-brain barrier is tempered by the detection limit of the GC/MS method used in that study and the elimination half-life of pravastatin. The detection limit of the statins was 0.5 ng/ml. Lovastatin acid which has a partition coefficient of 1.51 as compared with pravastatin with a partition coefficient of −0.47 14 showed a peak drug level of 1.3 ng/ml in CSF, 2.5 h following drug administration. Certainly based on the differences in the partition coefficients of the two statins, it would be expected that higher levels of lovastatin acid would be detected than pravastatin but the level of lovastatin acid detected was near the detection limit of the assay. The GC/MS assay method may not have had the sensitivity to detect pravastatin. Further impeding pravastatin detection is that the elimination half-life of pravastatin in serum has been reported to be 1.8 h 15 and the first determination of pravastatin in CSF was 1h after drug administration. In the same study, it was also observed that there was a 30% reduction in lovastatin acid levels between 2.5 and 4.0 h following drug administration. The elimination half-life of lovastatin in serum is approximately 3 h 15. It has not been determined if statins are metabolized in the CNS. Alternatively, the reduction in lovastatin acid might due to active transport out of the CNS which could involve brain acid transporters and is discussed later in this section. Transport of statins into brain was determined in another study using the uptake of radiolabeled statins and an *in situ* rat brain perfusion technique 16. Permeability of statins into brain was strongly associated with drug lipophilicity and pravastatin permeability was similar to sucrose and it was concluded that pravastatin did not cross the blood-brain barrier 16. Uptake time points were 10, 20 and 30 seconds which may have been too early following drug administration. In the same study, it was shown that pravastatin partitions through brain capillary endothelial cells over a 10 minute incubation period *in vitro* albeit at a slower rate as compared with simvastatin and lovastatin.

Two recent studies report unequivocally that pravastatin can be detected in brain 17,18. As part of a study on statins and brain gene expression in mice, we quantified statin levels in brain and found that simvastatin, lovastatin and pravastatin could be detected in cerebral cortex of mice administered the drugs *in vivo* 17. Initially, we determined simvastatin, lovastatin and pravastatin levels in the cerebral cortex of mice who received statins for 21 days. Mice were sacrificed 24 h after the last statin administration, cerebral cortex dissected and statin levels were determined by LC-MS/MS. Relatively low levels of simvastatin and lovastatin lactones were detected but pravastatin was not detected in the cerebral cortex 24 h after the last drug administration. The low levels of simvastatin and lovastatin and the absence of pravastatin at 24 h were puzzling because of the significant effects of all 3 statins on brain gene expression. To begin to examine the possibility that statins did not accumulate in brain, we examined statins levels in mice given a single administration and then sacrificed at 1, 3, and 6 h following drug administration. Simvastatin, lovastatin and pravastatin were all detected in the cerebral cortex. Not surprisingly, brain levels of the three statins reflected their lipophilicity with simvastatin levels >lovastatin levels> pravastatin levels. The maximum average concentration of pravastatin in the cerebral cortex was 100 pmol/g and these levels are above the reported 50% inhibitory concentration (IC50) on membrane bound HMG-CoA reductase. Between 1 and 6 h there was a large reduction in statin levels and is

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similar to the reduction in lovastatin levels in the CSF study 13 discussed earlier in this review. Statins in brain would appear to be rapidly eliminated but the mechanism for the reduction has not been determined. Since those initial findings on statin levels in brain, a second paper reported levels of simvastatin and pravastatin in mouse brain homogenates after a 3 day drug treatment 18.

A general conclusion of the four studies discussed above is that lipophilic statins can easily cross the blood-brain barrier. Consensus is not observed as to whether hydrophilic statins such as pravastatin can enter the brain. The two studies 13,16 which concluded that pravastatin did not cross the blood-brain barrier may have been limited with respect to assay sensitivity and the sampling times. On the other hand, the two studies 17,18 which found that pravastatin could be detected in brain used high drug concentrations (100mg/kg and 200 mg/kg per body weight) and sensitive LC-MS/MS methods to quantify drug levels at pmol concentrations. Membrane partitioning of pravastatin and probably other statins administered as acids such as atorvastatin most likely does not explain how a statin acid enters the brain. However, pravastatin has been shown to be transported into cells by organic anionic transporters (OATs) and these transporters are expressed in brain 19–21. Another transporter that shuttles statin acids into cells is the monocarboxylic acid transporter 22. Uptake of lovastatin and simvastatin lactones occurrs by simple diffusion but the acid form of the two statins is taken up by means of a transport mechanism for monocarboxylic acid in bovine brain capillary endothelial cells 22. We found that simvastatin, lovastatin and pravastatin significantly increased expression of the gene that encodes the monocarboxylic acid transporter in brain tissue of mice chronically administered those drugs 17. An additional mechanism which could potentially facilitate movement of statins into brain is statin lactonization. Atorvastatin acid as well as simvastatin acid can undergo lactonization and thus could increase the availability of the lactone forms particularly atorvastatin which then could cross the blood-brain barrier 23. While atorvastatin is administered as an acid it undergoes lactonization *in vivo* 23. Serum levels of atorvastatin lactone and acid were comparable in human subjects 24. The partition coefficients of atorvastatin lactone (4.2) and simvastatin lactone (4.4) 14 are quite similar and it is predicted that atorvastatin lactone would indeed cross the blood-brain barrier.

CSF and brain levels of statins showed a marked decline within a few hours after drug administration. It is not known if statins are metabolized in brain. Simvastatin, atorvastatin and lovastatin are substrates for CYP3A4 which is the primary CYP isoform involved in the metabolism of those drugs 15 and this enzyme is present in rodent brain 25. There are no published data on whether statins induce CYP3A4 in brain. Pravastatin is not metabolized by CYP3A4 and it is eliminated mostly unchanged 15. A different mechanism which could contribute to the elimination of statins from brain is the transporter P-glycoprotein (P-gp). Previous studies have shown that P-gp is involved in the excretion, absorption and distribution of hydrophobic and amphipathic drugs 26. Statins are substrates for P-gp and this transporter may play a role in statin efflux from cells 27,28. P-gp is a major component of the blood-brain-barrier 29 and it is located in different brain cell types including neurons 28. P-gp could act as a transporter both at the blood-brain barrier and neuronal cells.

Statin-Induced Neuroprotection Versus Cell Death

In studies using animal models and various cell types, statins have been shown to provide protection to various types of insults, e.g., NMDA and glutamate 30,31, amyloid beta-protein 32, oxygen glucose deprivation 33,34, sodium nitroprusside 35 and bone morphogenetic protein 36. However, there are data indicating that statins can have toxic effects on brain neural cells, e.g., 37–42. In general, what distinguishes the studies on protective effects of statins versus toxic effects are the drug concentrations used in the

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different studies. Most studies reporting neuroprotection have used statin concentrations ranging between 100 nM and 1 μM although it was reported that rosuvastatin at a concentration of 5 μM reduced toxicity of oxygen-glucose deprivation in rat primary cortical neurons 34. Effects of rosuvastatin alone on cell viability were not reported in that study. Statin-induced reductions in cell viability have typically used drug concentrations ranging for example between 2 μM and 300 μM. An exception is a recent study which found that simvastatin at a concentration of 0.1 μM and 1 μM induced cell death in a mouse cerebellar slice culture preparation 43. Several issues are raised by this interesting study. One possibility is that statin effects differ in cerebellar tissue as compared to other brain areas. Cholesterol abundance is less in mouse synaptic plasma membrane of the cerebellum as compared with membranes of the cerebral cortex and hippocampus 44. Statin-induced reduction of cholesterol may have more pronounced effects in brain regions were cholesterol levels are lower relative to other brain regions. Also, there are no available data on comparisons between effects of statins on brain slices from postnatal mice versus primary neurons from embryonic mice which may have an influence of the actions of statins. A prudent observation is that the use of high statin concentrations in neural cells is unwarranted unless the goal is to induce cell dysfunction and death.

Even though statins are shown to be associated with neuroprotection both *in vitro* and *in vivo*, the issue of drug concentration should also be considered with respect to statin dosage used when treating human patients. We have shown that chronic administration of simvastatin (50 mg/kg/day for 21 days) in mice and guinea pigs 17,35 increased gene expression and protein levels of Bcl-2, the major anti-apoptotic gene/protein member of the Bcl-2 family. Both lovastatin (20 mg/kg/day i.p. for 14 days) and atorvastatin (20 mg/kg/day orally for 7 or 21 days) administered to rats reduced the proinflammatory calcitonin gene-related peptide and substance P in the dorsal root ganglion 36. The statin concentrations used in those three studies are noticeably higher than doses taken by human subjects which generally range between 5 and 80 mg tablet per day 45. The human dosage is not based on body weight. If statins however were prescribed for human patients based on body weight, a 70 kg human male would be taking approximately 0.07/mg/kg/day to 1.14/mg/kg/day which is far less than statin concentrations used in animal studies. The issue of statin concentration also applies to the *in vitro* studies where relatively low drug concentrations (0.1 to 1 μM) were employed. Brain statin concentrations were found to be at the pmol level in brain tissue of mice administered different statins 17,18 and those *in vivo* levels are distinctly lower than the statin concentrations used in studies of neuroprotection in neural cells. It is worth noting that there are no data on intracellular statin concentrations of drug treated cells and is further evidence of the lack of basic data on statins.

Statins and Brain Isoprenoids

The mevalonate pathway produces several biologically active molecules which play an important role in cell function. One of those molecules is cholesterol which has certainly garnered intensive study. Upstream of cholesterol in that pathway are two isoprenoids farnesyl pyrophosphate (FPP) and geranylgeranyl pyrophosphate (GGPP) which are receiving increasing attention both with respect to normal function and their potential contributions to pathophysiology associated with coronary heart disease, cancer and neurodegenerative diseases 46–49. Until recently, the protective effects of statins in coronary heart disease were largely attributable to a reduction in cholesterol levels but there is emerging evidence that the reduction of isoprenoid levels specifically FPP and GGPP may also be important in preventing cardiovascular disease by reducing the abundance of prenylated proteins 46. FPP and GGPP belong to a group of isoprenoids which include for example, ubiquinone and dolichol. FPP is a C_{15} isoprenoid which is the key branch point in the synthesis of ubiquinone, dolichol and GGPP as well as cholesterol. In addition to its

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critical role in synthesis, FPP posttranslationally prenylates by farnesyl transferase the small GTPases such as H-ras, Rheb, and other proteins containing a cysteine in the C-terminal CaaX box (C = cysteine, a = aliphatic amino acid and X = C-terminal amino acid) [50,51]. FPP is a substrate for geranylgeranyl pyrophosphate synthase resulting in the production of GGPP, a 20-carbon lipid as compared with the 15-carbon FPP. Whereas FPP prenylates proteins containing a cystine of the CaaX box of the C terminal, GGPP on the other hand prenylates proteins containing a C-terminal leucine residue by geranylgeranyl prenyltranferase-1 (GGTase-1) such as the Rho family of GTPase and Rac-1. These and other prenylated proteins are integral components of complex signaling networks and control diverse cellular activities including intracellular vesicle transport, cell adhesion, endocytosis, cytoskeletal organization, receptor signaling, cell cycle progression and gene expression [51].

It has been suggested that the neuroprotective effects of statins may be due in part to a reduction in FPP and/or GGPP levels which in turn lessens abundance of prenylated proteins and which may or may not be cholesterol independent [5,9,48]. To rule out the contribution of statin-induced lowering of cholesterol levels, one approach is to incubate cells with a squalene synthase inhibitor which prevents the production of squalene and subsequently cholesterol. The reasoning is that reducing cholesterol levels by non-statin intervention leaves FPP and GGPP levels unaffected. If reducing cholesterol levels by squalene synthase inhibition has the same effect, such as increasing protein A as did a statin, then the conclusion is that the effects are cholesterol-dependent. On the other hand, if there is no change in the effect of non-statin cholesterol reduction on protein A then the conclusion is that such effects may be mediated by statin-induced reduction of FPP or GGPP. To test those possibilities, two different approaches can be used: 1) incubating cells with exogenous FPP or GGPP in the presence of a statin; or 2) incubating cells with specific inhibitors of FPP and GGPP transferases involved in protein prenylation. If the addition of FPP or GGPP reduces effects of the statin on protein A, then one interpretation is that the statin effects are due to a reduction of FPP or GGPP levels but that has never been directly tested in brain, i.e., FPP and GGPP levels determined. There are no *in vivo* or *in vitro* data on effects of statins on FPP and GGPP levels in brain. In the example of using an inhibitor of one of the transferases, if effects on response A are similar to the statin effects, then the conclusion is that statin-induced reduction of FPP or GGPP and not cholesterol is the underlying mechanism.

In practice, the manipulation of FPP, GGPP and cholesterol levels as described above seems a workable approach to tease out the contributions of the different mevalonate-derived lipids acted upon by statins. We and other groups have used such an approach [32,52,53] but it is problematic for several different reasons. In a typical experiment, μmol amounts or greater of FPP or GGPP are incubated with cells. Recent studies of mouse brain [54] and human brain [55] have demonstrated that levels of FPP and GGPP are in the pmol to nmol range and that GGPP abundance is markedly higher than FPP. Until those two studies, data on brain FPP and GGPP levels had not been determined and thus the rationale was not clear for incubating cells with μmol amounts of FPP and GGPP. In view of the finding that brain GGPP levels were several fold higher than FPP levels, adding μmol amounts of FPP to cells could possibly have confounding effects on regulation within the HMG-CoA reductase pathway. An example of unexpected consequences when perturbing a member of the HMG-CoA reductase pathway is a study which reported that cells treated with the squalene synthase inhibitor, zaragozic acid, showed over a 250-fold increase in FPP levels but only a 4-fold increase in GGPP levels in the mouse fibroblast cell line NIH3T3 [56]. The reasoning for reducing cholesterol levels by means which do not reduce FPP and GGPP levels allows one to determine if effects of statins are due to only cholesterol reduction. However, the key observation that zaragozic acid treatment of cells has such a remarkable impact on FPP

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levels belies the notion that only cholesterol levels are altered by such treatment. That experiment was a relatively uncomplicated experiment in design but the fact that FPP and GGPP levels were quantified provided major insight into FPP and GGPP homeostasis and such insight is lacking in understanding isoprenoid regulation in brain and underscores the need for more research. To date, there are no published data effects of statins *in vivo* or *in vitro* on brain FPP and GGPP levels.

CONCLUSIONS

Statins may be efficacious in preventing and treating certain neurodegenerative diseases and support is based in part on animal and cell culture studies. Further progress however in understanding the mechanisms of statin-induced neuroprotection are hampered by a lack of fundamental data on actions of statins in the CNS. Three major topics which need to be addressed are the pharmacokinetics of lipophilic and hydrophilic statins in brain, cell protection versus cell death and regulation of brain isoprenoids. There is inadequate knowledge on each of these topics which we suggest has in some instances resulted in erroneous conclusions. For example, the notion that hydrophilic statins do not cross the blood-brain barrier is not supported by the limited data available. Basic studies are needed on statin transport in and out of brain as well as whether metabolism of statins occurs in the CNS. The issue of statin-induced cell protection versus cell death may simply be explainable by drug concentration but this issue requires further investigation. Regulation of brain FPP and GGPP are not understood. It is assumed that FPP and GGPP levels are reduced in brains of animals and neural cells treated with statins but there are no reported data. What is instructive is that the two recent studies reporting brain levels of FPP and GGPP in mice and humans found that GGPP was in greater abundance than FPP and isoprenoid levels were in the range of pmol to nmol. From a practical standpoint, those data have relevance to the procedure of examining effects of statins by adding back μmol amounts of FPP and/or GGPP in neural cells. Statins may be neuroprotective but efforts toward understanding the potential mechanisms will be enhanced by advancements in the CNS pharmacokinetics and pharmacodynamics of statins.

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References

1. Tobert JA. Lovastatin and beyond: The history of the HMG-CoA reductase inhibitors. Nature Reviews. 2003; 2:517–526.
2. Law MR, et al. Quantifying effect of statins on low density lipoprotein cholesterol, ischaemic heart disease, and stroke: Systematic review and meta-analysis. Br. Med. J. 2003; 326:1–7.
3. Nassief A, Marsh JD. Statin Therapy for Stroke Prevention. Stroke. 2008; 39:1042–1048. [PubMed: 18258828]
4. Wahner AD, et al. Statin use and the risk of Parkinson disease. Neurology. 2008; 70:1418–1422. [PubMed: 18184918]
5. Eckert GP, et al. Statins: Drugs for Alzheimer's Disease? J. Neural Transm. 2005; 112:1057–1071. [PubMed: 15682268]
6. Neuhaus O, Hartung HP. Evaluation of atorvastatin and simvastatin for treatment of multiple sclerosis. Expert Rev Neurother. 2007; 7:547–556. [PubMed: 17492904]
7. Tapia-Perez JH, et al. Effect of Rosuvastatin on Amenesia and Disorientation after Traumatic Brain Injury (NCT003229758). J Neurotrauma. 2008; 25:1011–1017. [PubMed: 18690806]

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8. Stepien K, et al. Neuroprotective properties of statins. Pharmacol Rep. 2005; 57:561–569. [PubMed: 16227638]
9. Eckert GP, et al. Cholesterol-lowering drugs and Alzheimer's disease. Future Lipidol. 2007; 2:423–432.
10. Tapia-Perez H, et al. Statins and brain protection mechanisms. Revista de Neurologia. 2007; 45:359–364. [PubMed: 17899518]
11. Farooqui AA, et al. Comparison of biochemical effects of statins and fish oil in brain: The battle of the titans. Brain Res Rev. 2007; 56:443–471. [PubMed: 17959252]
12. Orr JD. Statins in the spectrum of neurologic disease. Curr Atheroscler rep. 2008; 10:11–18. [PubMed: 18366980]
13. Botti RE, et al. Concentrations of pravastatin and lovastatin in cerebrospinal fluid in healthy subjects. Clin. Neuropharmacol. 1991; 14:256–261. [PubMed: 1906375]
14. Ishigami M, et al. A comparison of the effects of 3-hydroxy-3-methylglutaryl-coenzyme A (HMG-CoA) reductase inhibitors on the CYP3A4-dependent oxidation of mexazolam in vitro. Drug Metab Dispos. 2001; 29:282–288. [PubMed: 11181496]
15. Schachter M. Chemical, pharmacokinetic and pharmacodynamic properties of statins: an update. Fund & Clin Pharmacol. 2004; 19:117–125.
16. Saheki A, et al. In vivo and in vitro blood-brain barrier transport of 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase inhibitors. Pharmaceutical Research. 1994; 11:305–311. [PubMed: 8165193]
17. Johnson-Anuna LN, et al. Chronic administration of statins alters multiple gene expression patterns in mouse cerebral cortex. J. Pharmacol. Exp. Ther. 2005; 312:786–793. [PubMed: 15358814]
18. Thelen KM, et al. Brain cholesterol synthesis in mice is affected by high dose of simvastatin but not of pravastatin. J Pharmacol Exp Ther. 2006; 316:1146–1152. [PubMed: 16282522]
19. Hasegawa M, et al. Contribution of organic anion transporters to the renal uptake of anionic compounds and nucleoside derivatives. J Pharmacol Exp Ther. 2003; 305:1087–1097. [PubMed: 12660303]
20. Takeda M, et al. Evidence for a role of human organic anion transporters in the muscular side effects of HMG-CoA reductase inhibitors European. Journal of Pharmacology. 2004; 483:133–138.
21. Kusuhara H, et al. Molecular cloning and characterization of a new multispecific organic anion transporter from rat brain. J. Biol. Chem. 1999; 274:13675–13680. [PubMed: 10224140]
22. Tsuji A, et al. Transport mechanism of 3-hydroxy-3-methylglutaryl conenzyme A reductase inhibitors at the blood-brain barrier. J. Pharmacol. Exp. Ther. 1993; 267:1085–1090. [PubMed: 8263769]
23. Jacobsen W, et al. Lactonization is the critical first step in the disposition of the 3-hydroxy-3-methylglutaryl-CoA reductase inhibitor atorvastatin. Drug Metab Dispos. 2000; 28:1369–1378. [PubMed: 11038166]
24. Kantola T, et al. Effect of itraconazole on the pharmacokinetics of atorvastatin. Clin Pharmacol Ther. 1998; 64:58–65. [PubMed: 9695720]
25. Pai HV, et al. Differential metabolism of alprazolam by liver and brain c (P4503A) to pharmacologically active metabolite. Pharmacogenomics. 2002; 2:243–258.
26. Schinkel AH. The physiological function of drug-transporting P-glycoproteins Semin. Cancer Biol. 1997; 8:161–170.
27. Bogman K, et al. HMG-CoA reductase inhibitors and P-glycoprotein modulation. Br. J. Pharmacol. 2001; 132:1183–1192. [PubMed: 11250868]
28. Hirrlinger J, et al. Expression of mRNAs of multidrug resistance proteins (Mrps) in cultured rat astrocytes, oligodendrocytes, microglial cells and neurons. J. Neurochem. 2002; 82:716–729. [PubMed: 12153495]
29. Sun H, et al. Drug efflux transporters in the CNS. Adv. Drug Deliv. Rev. 2003; 55:83–105. [PubMed: 12535575]
30. Zacco A, et al. 3-Hydroxy-3-methylglutaryl coenzyme A reductase inhibitors protect cortical neurons from excitotoxicity. J. Neurosci. 2003; 23:11104–11111. [PubMed: 14657168]

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31. Bösel J, et al. Neuroprotective effects of atorvastatin against glutamate-induced excitotoxicity in primary cortical neurones. J. Neurochem. 2005; 92:1386–1398. [PubMed: 15748157]
32. Johnson-Anuna LN, et al. Simvastatin protects neurons from cytotoxicity by up-regulating Bcl-2 mRNA and protein. J. Neurochem. 2007; 101:77–86. [PubMed: 17241114]
33. Lim JH, et al. Simvastatin prevents oxygen and glucose deprivation/reoxygenation-induced death of cortical neurons by reducing the production and toxicity of 4-hydroxy-2E-nonenal. J. Neurochem. 2006; 97:140–150. [PubMed: 16515553]
34. Domoki F, et al. Rosuvastatin induces delayed preconditioning against oxygen-glucose deprivation in cultured cortical neurons. Am J Physiol Cell Physiol. 2009; 296:C97–C105. [PubMed: 18971391]
35. Franke C, et al. Bcl-2 upregulation and neuroprotection in guinea pig brain following chronic simvastatin treatment. Neurobiology of Disease. 2007; 25:438–445. [PubMed: 17157514]
36. Bucelli RC, et al. Statins decrease expression of the proinflammatory neuropeptides calcitonin gene-related peptide and substance P in sensory neurons. J Pharmacol Exp Ther. 2008; 324:1172–1180. [PubMed: 18079356]
37. Michikawa M, Yanagisawa K. Inhibition of cholesterol production but not of nonsterol isoprenoid products induces neuronal cell death. J. Neurochem. 1999; 72:2278–2285. [PubMed: 10349836]
38. Tanaka T, et al. Geranylgeranyl-pyrophosphate, an isoprenoid of mevalonate cascade, is a critical compound for rat primary cultured cortical neurons to protect the cell death induced by 3-hydroxy-3-methylglutaryl-CoA reductase inhibition. J. Neurosci. 2000; 20:2852–2859. [PubMed: 10751437]
39. Schulz JG, et al. HMG-CoA reductase inhibition causes neurite loss by interfering with geranylgeranylpyrophosphate synthesis. J. Neurochem. 2004; 89:24–32. [PubMed: 15030386]
40. Jiang Z, et al. Lovastatin-induced up-regulation of the BH3-only protein, Bim, and cell death in glioblastoma cells. J. Neurochem. 2004; 89:168–178. [PubMed: 15030401]
41. März P, et al. Statins induce differentiation and cell death in neurons and astroglia. Glia. 2007; 55:1–12. [PubMed: 16998865]
42. Klopfleisch S, et al. Negative impact of statins on oligodendrocytes and myelin formation *in vitro* and *in vivo*. J. Neurosci. 2008; 28:13609–13614. [PubMed: 19074034]
43. Xiang Z, Reeves SA. Simvastatin induces cells death in a mouse cerebellar slice culture (CSC) model of developmental myelination. Exp. Neurol. 2009; 215:41–47. [PubMed: 18929563]
44. Chochina SV, et al. Amyloid β-peptide$_{1-40}$ increases neuronal membrane fluidity: Role of cholesterol and brain region. J. Lipid Res. 2001; 42:1292–1297. [PubMed: 11483631]
45. Solomon KR, Freeman MR. Do the cholesterol-lowering properties of statins affect cancer risk? Trends in Endocrinology and Metabolism. 2008; 19:113–121. [PubMed: 18356074]
46. Liao JK, Laufs U. Pleiotropic effects of statins. Annu. Rev. Pharmacol. Toxicol. 2004; 45:89–118. [PubMed: 15822172]
47. Swanson KM, Hohl RJ. Anti-cancer therapy: targeting the mevalonate pathway. Curr Cancer Drug Targets. 2006; 6:15–37. [PubMed: 16475974]
48. Cole SL, Vassar R. Isoprenoids and Alzheimer's disease: a complex relationship. Neurobiology of Disease. 2006; 22:209–222. [PubMed: 16406223]
49. Wood, WG., et al. Cholesterol-A Janus-Faced Molecule in the Central Nervous System. In: lajtha, A.; Reith, MEA., editors. Handbook of Neurochemistry and Molecular Neurobiology-Neural Membranes and Transport. New York: Springer; 2007. p. 151-170.
50. Basso AD, et al. Farnesyl transferase inhibitors. J. Lipid Res. 2006; 47:15–31. [PubMed: 16278491]
51. MC Taggart SJ. Isoprenylated proteins. Cell Mol Life Sci. 2006; 63:255–267. [PubMed: 16378247]
52. Cole SL. Statins cause intracellular accumulation of amyloid precursor protein. β-secretase-cleaved fragments, and amyloid β-peptide via an isoprenoid-dependent mechanism. J. Biol. Chem. 2005; 280:18755–18770. [PubMed: 15718241]
53. Ostrowski SM, et al. Statins reduce amyloid-β production through inhibition of protein isoprenylation. J. Biol. Chem. 2007; 282:26832–26844. [PubMed: 17646164]

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54. Tong H, et al. Quantitative determination of farnesyl and geranylgeranyl diphosphate levels in mammalian tissue. Anal Biochem. 2008; 378:138–143. [PubMed: 18457649]
55. Hooff GP, et al. Isoprenoid quantitation in human brain tissue: a validated HPLC-fluorescence detection method for endogenous farnesyl- (FPP) and geranylgeranylpyrophosphate (GGPP). Anal Bioanal Chem. 2008; 392:673–680. [PubMed: 18690423]
56. Tong H, et al. Simultaneous determination of farnesyl and geranylgeranyl pyrophosphate levels in cultured cells. Anal Biochem. 2005; 336:51–59. [PubMed: 15582558]