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Cell type-specific transcriptome profiling in mammalian brains

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1. ABSTRACT

A mammalian brain contains numerous types of cells. Advances in neuroscience in the past decade allow us to identify and isolate neural cells of interest from mammalian brains. Recent developments in high-throughput technologies, such as microarrays and next-generation sequencing (NGS), provide detailed information on gene expression in pooled cells on a genomic scale. As a result, many novel genes have been found critical in cell type-specific transcriptional regulation. These differentially expressed genes can be used as molecular signatures, unique to a particular class of neural cells. Use of this gene expression-based approach can further differentiate neural cell types into subtypes, potentially linking some of them with neurological diseases. In this article, experimental techniques used to purify neural cells are described, followed by a review on recent microarray- or NGS-based transcriptomic studies of common neural cell types. The future prospects of cell type-specific research are also discussed.

2. INTRODUCTION

The brain is one of the most complex and important organs in a mammal's body. A typical mammalian brain contains 10^8 (mouse) to 10^{11} (human) neurons and even larger numbers of glia (1). It has been the subject of a great deal of research, due to its importance with respect to behavior, perception, thought, and emotion. It is also the root of a number of serious diseases, including dementia, epilepsy, strokes, headache disorders, Parkinson's disease, and Alzheimer's disease. The World Health Organization (WHO) published a report in 2007 estimating that up to 1 billion people, or one in six of the world's population, suffer from neurological disorders (2). This indicates that these diseases pose a great threat to public health. Responding to this threat, the United States has launched the Brain Research through Advancing Innovative Neurotechnologies (BRAIN) Initiative to develop new tools and technologies for deepening our understanding of the brain (3-5).

Neural tissues in mammals include a large number of cell types. Although much progress has been made in the development of techniques to identify common neural cell types, the total number of neural cell types and subtypes is still far from clear. As such, one of the priority research areas of the BRAIN Initiative is to differentiate neural cell types and determine their roles in health and disease (5). Successful implementation of the Initiative would facilitate a better understanding of various neurological diseases, aiding in their diagnosis and treatment.

High-throughput technologies, including microarrays (6, 7) and next-generation sequencing (NGS) (8, 9), have helped dissect numerous neurological diseases (reviewed in ref. 10-12) and allow brain functional

annotations at different levels. For instance, at the level of brain regions (*e.g.,* prefrontal area), several studies have provided a comprehensive atlas of gene expression across the brain (13-15). At the level of single cells, expression profiling of tens of thousands of genes (16-19) has been achieved using multiplex PCR (20). However, the brain-wide data are difficult to interpret because the information is not available for the localization of individual transcripts at the cellular level (21, 22), whereas the single-cell methods have issues of increased false negatives and reduced reproducibility (23, 24). Given these difficulties, understanding of gene expression at the cellular level mainly comes from pooled cells obtained by several techniques such as laser-capture microdissection (LCM) (25-27), fluorescence-activated cell sorting (FACS) (28-32), immunopanning (PAN) (32-34), and translating ribosome affinity purification (TRAP) (35-38).

In this article, we will first provide an overview of common cell types in mammalian brains and of experimental techniques for cell purification and identification. Then, we will review recent microarray- and NGS-based studies on transcriptomics of specific neural cell type. The future prospects of cell type-specific studies are also discussed.

3. IDENTIFICATION AND ISOLATION OF CELL-TYPE SPECIFIC POPULATIONS IN A MAMMALIAN BRAIN

3.1. Common neural cell types

A mammalian brain is made up of a large number of cell types that are vital to proper functioning of the central nervous system (CNS). Foremost among them is neuron, a primary vehicle for long-distance electrical communication and computation among cells in mammals (39). Neurons are interconnected cells, each possessing a large cell body (soma), as well as cell projections called dendrites and an axon. The dendrites are thin, branched projections that receive neurotransmitters from other neurons, while the axon is a long projection sending electrical signals to the next neuron. Neurons send signals among themselves by changing electrical potentials, which can spread along the axon of a neuron. The bulb-like end of the axon, termed axon terminal, is separated from the dendrites of the next neuron by a narrow space (synapse). When electrical signals travel to the axon terminal, neurotransmitters are released across synapse and bind post-synaptic receptors, stimulating receiving neurons to modulate electrical potentials and continue nerve impulse. Neurons are arguably the most important cell type in the body, enabling computations required for vital behaviors such as balance, communication, and the ability to learn and make decisions. Changes in the gene expression of neurons can lead to an over- or under-expression of important genes, which can radically change the overall topology of the CNS and lead to severe disorders. For example, the Parkinson's disease is characterized by an accumulation of alpha-synuclein and a subsequent deficiency of dopamine in the brain as the dopamine-producing cells die (40). The Huntington's disease is caused by the production and accumulation of mutant Huntingtin (Htt) proteins (41).

Glial cells are non-neuronal cells, including astrocytes, oligodendrocytes, and microglia, which are smaller in size (compared with neurons) and vary in structure (42). Astrocytes are star-shaped cells with great structural complexity (43-45). Traditionally, astrocytes are considered to be ancillary, satellite cells that provide a physical support network to neurons, and regulate the environment so that neurons can function properly. As such, astrocytes maintain extracellular ion balance and pH homeostasis (46). They hold important stores of glycogen, providing surrounding cells with glucose as needed (47). Moreover, astrocytes interact with the synapses of neurons, and work to both produce and remove neurotransmitters and other compounds from the intercellular space (48). Recent studies have revealed new roles of astrocytes (49-52). First, astrocytes establish separate territories that define functional domains of a brain (49). Second, astrocytes can release neuroactive agents such as glutamate to modulate synaptic transmission (50). Third, astrocytes interact with neurons and endothelial cells to form higher-order gliovascular units, bridging neuronal and vascular structure to match local neural activity and blood flow (51). Currently, astrocytes are seen as an important communication element of the brain, and their dysfunction may lead to aberrations of neuronal circuitry that underlies several neurodevelopmental disorders such as the Rett syndrome (53) and the fragile X mental retardation (54) (reviewed in ref. 55, 56).

Besides astrocytes, oligodendrocytes and microglia are two types of glial cells. The main function of oligodendrocytes is to produce and maintain myelin sheath which wraps around axons. Myelin helps to support and insulate axons in the CNS. Oligodendrocyte progenitor cells (OPCs) are precursors to oligodendrocytes and can also differentiate into neurons and astrocytes. Microglial cells are resident macrophages that provide the first line of immune defense in the CNS. Oligodendrocytes and microglial cells account for 75.6.% and 6.5.% of the cerebral cortex glia, respectively (57).

3.2. Experimental techniques to isolate and purify specific neural cell types

Cell type-specific transcriptomics require completion of several steps (Figure 1). First, brain tissues need to be isolated as fixed/frozen sections or live samples. Second, cells of interest need to be identified and collected. Third, RNA from the collected cells needs to be extracted and, if necessary, amplified. Fourth, relative mRNA expression levels need to be measured

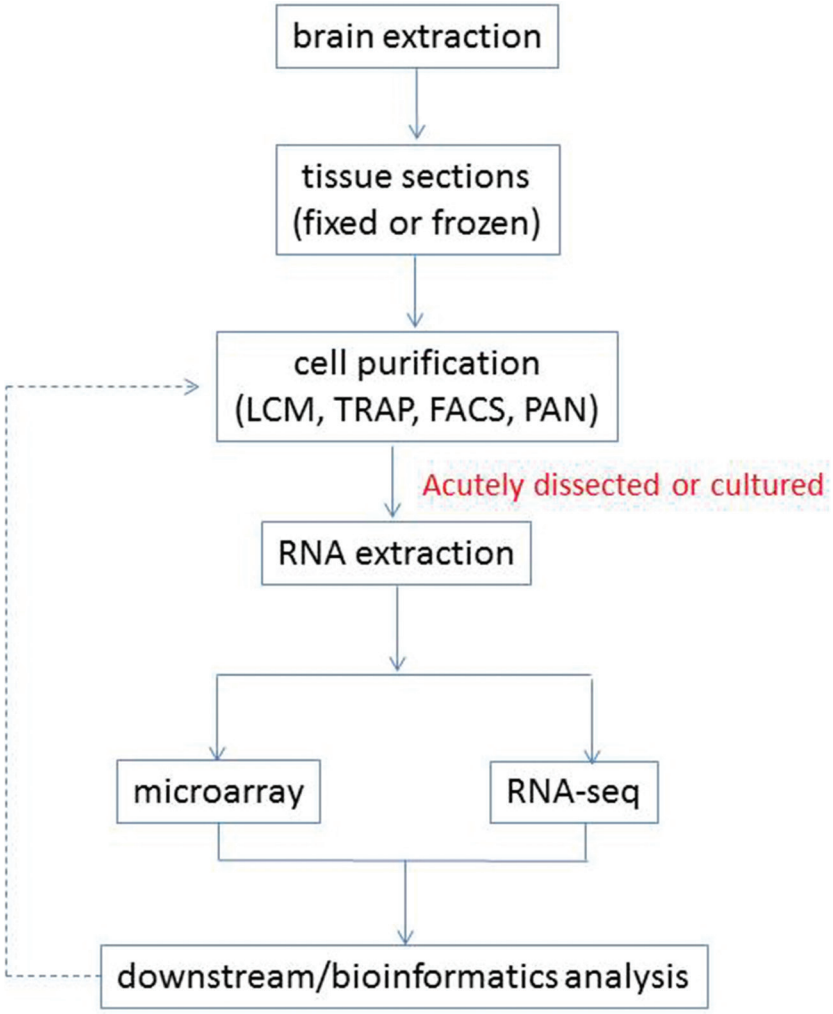

Figure 1. Typical workflow of cell type-specific transcriptome profiling experiments. Cell-specific transcriptome profiling requires completing several steps such as brain sample collection, cell purification, RNA extraction, and RNA abundance measurement by microarray or RNA sequencing. At the last step, genes that are differentially expressed are identified by bioinformatic tools. If the protein products of these genes are directed to the surface of target cells, they can be used as cell markers. The cells presenting these proteins on the surface can be identified and purified by FACS and PAN. This process can undergo multiple iterations to differentiate a neural cell type into many subclasses.

by microarray or RNA-seq. Finally, downstream analyses need to be performed using bioinformatics tools. In this section, we focus on the first two steps because tissue heterogeneity of a mammalian brain has been a major obstacle (Figure 1). A pure collection of neural cells is the key to profiling cell-type specific transcriptomes. It is worth noting that the problem of neural cell identification has been discussed (58, 59). Cell type-specific markers and Cre-driver mouse lines are available to identify specific neural cell types (see online resources in ref. 60).

Technically, specific neural cell types can be harvested by four methods: (1) laser-capture microdissection (LCM) or laser-directed microdissection (LDM) (25-27); (2) fluorescence-activated cell sorting (FACS) (28-32); (3) immunopanning (PAN) (32-34); and (4) translating ribosome affinity purification (TRAP) (35-38). A quantitative analysis on cell type-specific microarray data has revealed that LCM and TRAP samples show significantly higher levels of contamination than FACS and PAN samples (60). Below, we provide a brief overview of these four methods.

3.2.1. LCM/LDM

LCM and LDM use a laser to isolate specific cell populations under a microscope from mounted thin-tissue sections that are either fixed or frozen. In LCM, a laser excises a small region (~7.5. μm) of a plastic membrane on the surface of tissue sections. Cells underneath adhere to the membrane upon cooling and are collected after the membrane is removed. One limitation of this method is that it does not allow users to extract a given cell by tracing its particular morphology. To overcome this limitation, LDM uses a much narrower ultraviolet laser

(~0.5. μm), which allows users to make precise cuttings along the outline of target cells.

3.2.2. FACS

FACS requires that target cells are labelled by fluorophores. The fluorophores are typically attached to antibodies that recognize a target feature on the cells. Based on the specific light scattering and fluorescent characteristics of each cell, FACS can sort a heterogeneous mixture of a cell population into subgroups that may belong to specific cell types. In FACS, the live, acutely-dissected brain tissue is digested in a protease solution with artificial CSF (ACSF), which keeps dissociated cells in a healthy condition. One main advantage of the FACS method is that it can sort a large number of cells in a high-throughput manner.

3.2.3. PAN

PAN relies on antibodies against specific proteins on the surface of target cells, not fluorescent signals. Panning plates are first coated with antibodies, and dissociated cells are then placed in the plates for 30 minutes to 1 hour, allowing target cells to bind the antibodies. The plates are then washed to collect the cells of interest. Multiple iterations of plating and antibody interactions may be required, which may be more time-consuming than other techniques.

3.2.4. TRAP

TRAP uses special transgenic mice with restricted cell populations in the CNS. TRAP harvests RNA on labelled polysomes directly from tissue homogenates. Only ribosome-associated mRNA rather than the full population of transcribed RNA is detected. As a result, the noncoding RNAs that play an important role in gene regulation are discarded. Since tissue homogenates are used for analysis, TRAP samples often have contamination, as shown by previous studies (61).

4. CELL TYPE-SPECIFIC TRANSCRIPTOME PROFILING

4.1. Expression profiling of acutely-isolated cells

Microarrays have been used to analyze the functional genomics of different cell types acutely purified from the brain (Table 1). One of the first examples came from Dugas *et al.* (62) who compared gene expression in the oligodendrocytes (OLs) generated from cultured oligodendrocyte progenitor cells (OPCs) *in vitro* and the OLs isolated acutely from animal brains. The OLs and OPCs were purified by PAN. Dugas *et al.* (62) found that OL differentiation occurs in at least two sequential stages, the early stage and the late stage, which are characterized by different expression patterns of transcription factors and myelin genes. Genes encoding cytoskeletal proteins are up-regulated during the OL differentiation. These findings were confirmed later by Cahoy *et al.* (32) who found that multiple signaling pathways including actin cytoskeleton signaling are enriched in the OLs. A separate study showed that a miRNA species, miR-9, is important for the OL differentiation and its expression inversely correlates with the expression of peripheral myelin protein PMP22 (64). This finding highlights the importance of miRNAs in neuronal cell specification (75).

Transcriptomic analyses of pooled neurons have shown that neurons have an elevated expression of genes involved in glycolysis and oxidative metabolism (63). The enzymes in the tricarboxylic acid (TCA) cycle are expressed at low levels. Several pathways involved in calcium signaling, axonal guidance signaling, glutamate receptor signaling, and GABA receptor signaling are enriched in neurons (32). Further studies on transcriptomes of rostral and caudal serotonin neurons provide evidence for the complexity of gene regulatory networks in different types of neurons (66). In particular, hundreds of transcripts are differentially expressed in rostral and caudal serotonin neurons, in which a homeodomain code seems to play a key role in differentiating these two types of neurons. Finally, gene expression profiling of neural stem cells (NSCs) has revealed that the growth factor insulin-like growth factor 2 (IGF2) is expressed at high levels, which suggests that IGF2 plays an important role in adult neurogenesis (68).

Expression profiling on isolated astrocytes has uncovered that the enzymes in the tricarboxylic acid (TCA) cycle are expressed at higher levels than in neurons (61). Not surprisingly, the TCA cycle is found to be one of the metabolic pathways enriched in astrocytes (32). Moreover, the Notch signaling pathway is one of the top pathways enriched in astrocytes. Although Notch signaling has been suggested to play a role in differentiating neural progenitor cells into astrocytes, these findings indicate that Notch signaling may be required for maintaining astrocyte fate, preventing them from reverting to undifferentiated states (76). Note that gene expression patterns in astrocytes vary as a function of age: young astrocytes have high expression levels of genes involved in neuronal differentiation and hemoglobin synthesis, whereas aged astrocytes are characterized by increased inflammatory phenotypes and zinc ion binding (70).

Transcriptomic analyses of purified microglia reveal distinct gene expression patterns for young and aged microglia. Young microglia cells are characterized with increased transcript levels of chemokines such as Ccl2 and Ccl7 (70). These chemokines have been linked to differentiation and maturation of neurons (77). By contrast, genes within the tumor necrosis factor-ligand family, such as *Tnfsf12* and *Tnfsf13b*, are up-regulated in aged microglia (70, 78, 79). Microarray analyses on acutely-isolated brain endothelial cells (65) and Purkinje cells (67) have also provided cell type-specific gene signatures.

Table 1. Cell type-specific transcriptomic studies in mammalian brain

Cell preparation	Purification	Cell type	Exp. method	References
Acutely purified	PAN	Oligodendrocyte	Microarray	62
Acutely purified	LDM	Neuron	Microarray	26
Acutely purified	FACS	Astrocyte	Microarray	63
Acutely purified	TRAP	24 Cell types	Microarray	35
Acutely purified	FACS, PAN	Astrocyte, neuron, oligodendrocyte	Microarray	32
Acutely purified	FACS	Oligodendrocyte	Microarray	64
Acutely purified	FACS	Endothelial cell	Microarray	65
Acutely purified	FACS	5HT neuron	Microarray	66
Acutely purified	LCM	Purkinje cell	Microarray	67
Acutely purified	FACS	Neural stem cell	Microarray	68
Acutely purified	FACS	Microglia	Microarray	69
Acutely purified	FACS	Astrocyte, microglia	Microarray	70
Acutely purified	FACS	Microglia	RNA-seq	71
Acutely purified	LCM	Neuron	RNA-seq	72
Acutely purified	FACS, PAN	Astrocyte, neuron, oligodendrocyte, endothelial cell, microglia, pericyte	RNA-seq	73
Cultured		Oligodendrocyte	Microarray	62
Cultured		Astroglia	Microarray	32
Cultured		Microglia	Microarray	69
Cultured		Neuron	RNA-seq	74
Cultured		Astrocyte, neuron, oligodendrocyte progenitor cell	RNA-seq	92

The aforementioned gene expression profiling experiments are performed by microarrays. While microarray-based methods have been used for many years, these methods have numerous limitations. For example, microarrays can have cross-hybridization artifacts, detection difficulties due to the dye, and can be very limited in terms of alternative splicing (80). In recent years, next-generation sequencing (NGS) has become a popular tool for accurate, reproducible measurements of transcriptomics. This technique can provide sequences of all RNA molecules present within a cell, allowing for an accurate counting of different RNA species. RNA samples can be prepared from pooled cells, brain regions, or even individual cells (81).

Due to its unique ability to uncover details on both the expression level and isoform diversity, RNA sequencing (RNA-seq) has been used to identify gene expression signatures and gene splicing in acutely-isolated neurons (72) and microglia (71). Recently, Zhang *et al.* (73) used RNA-seq to generate transcriptome databases for eight cell types including neurons, astrocytes, oligodendrocyte precursor cells, newly formed oligodendrocytes, myelinating oligodendrocytes, microglia, endothelial cells, and pericytes from the mouse cerebral cortex. Remarkably, they found that the majority (~92%) of differentially expressed genes identified by microarray (32) are found by RNA-seq. As expected, the authors uncovered well-known cell type-specific markers, e.g., Aqp4 and Aldh1l1 for astrocytes, and Dlx1 and Stmn2 for neurons (32, 62, 65, 69, 71). Moreover, they have detected a large number of new genes with previously unknown cell type-specific distributions, highlighting the improved sensitivity of RNA-seq over microarrays. For example, the autism and schizophrenia-associated gene *Tspan7* is enriched in astrocytes, whereas a gene encoding a novel transmembrane protein Tmem59l is enriched in neurons. These data have provided a set of cell type-specific transcription factors that are important for cell fate determination and differentiation. These databases also allow the detection of alternative splicing events in glia, neurons, and vascular cells of the brain. One important finding is that *PKM2*, the gene encoding the glycolytic enzyme pyruvate kinase, has unique splicing forms in neurons and astrocytes. This may explain how neurons and astrocytes differ in their ability to regulate the glycolytic flux and lactate generation (82-86).

4.2. Expression profiling of cultured cells

Before the development of technologies like LCM/LDM, FACS, PAN, and TRAP, primary cultures of neural cells such as astroglia (87) had served as

an *in vitro* proxy for studying *in vivo* astrocytes. These cultured astrocytes have phenotypic characteristics that are significantly different from their *in situ* counterparts. For instance, astrocytes *in situ* are highly polarized cells, with distinct sets of processes that project to either synapses or vascular walls (88, 89). Cultured astrocytes, however, appear non-polarized with an epithelioid-like shape in the cultures. Several studies have found that genes that are induced in the cultured astrocytes are not necessarily expressed *in vivo*, suggesting that cultured astroglia do not represent the same cell type as *in vivo* astrocytes (32, 90). This is not true however for oligodendrocytes and retinal ganglion cells (see below), suggesting that at least for certain neural cell types, cells cultured *in vitro* mimic those *in vivo*. Understanding the differences in gene expression between cells grown *in vitro* and those acutely purified from animal brains is critical for making the right decision on what cells can be used under which situation.

Dugas *et al.* have compared the transcriptomic profiles between cultured OLs and acutely-purified OLs, and found a remarkable similarity in gene expression between the two groups (62). This result indicates that normal OL differentiation can take place in the absence of heterologous cell-cell interactions. This similarity between cultured and acutely-isolated cells is also observed for retinal ganglion cells (91), but not for astrocytes (32) and microglia (69).

Recently, we conducted a comprehensive analysis of transcriptomes in cultured neurons, astrocytes and OPCs through RNA-seq, and identified cell-specific marker genes and characteristic pathways that are known for these cell types (92). We compared our RNA-seq data with those from Zhang *et al*. (73) and found a number of genes are differentially expressed in cultured cells compared to acutely isolated cells. We conclude that the findings obtained from cells cultured *in vitro* should not be extrapolated to cells *in vivo*, especially when targeting genes or pathways associated with neurological diseases.

5. CONCLUSIONS AND FUTURE PROSPECTS

During the past decade, various types of neural cells have been identified and isolated by experimental techniques such as LCM, FACS, PAN, and TRAP. Most of these techniques depend on antibodies recognizing a small number of cell type-specific surface markers. Transcriptomic studies on the isolated cells of interest (Table 1) have identified numerous cell type-specific genes and pathways. Some of these genes can be used as molecular signatures to identify subclasses of neural cells. That is, if the protein products of these genes are directed to cell surface, they can be recognized by antibodies. A subclass of cells with these surface proteins can be isolated by FACS or PAN. This gene expression-based method allows us to identify many subtypes of a given neural population. This information may be very useful if we intend to link a neurological disease with a particular class of neurons. This gene expression-based classification of neural cells is a promising approach for cell type-specific research in the future.

Most cell type-specific studies so far are focused on transcriptomics, aiming to elucidate gene expression patterns of a given cell type. Few studies are dedicated to epigenomics. Analyses of genome-wide chromatin organization, including histone modification and DNA methylation, may shed new light on: (1) novel cell type-specific markers, (2) chromatin accessibility during differentiation, and (3) mechanisms underlying differential gene expression observed.

Finally, most studies to date have used cells from normal subjects. It would be more valuable to extend cell type-specific research to disease models. Understanding gene regulatory networks in the cell types responsible for a neurological disease would help to uncover the genetic and metabolic basis of this disease, which in turn, could open up new ways to diagnosis and new treatments of the disease. It would be an invaluable contribution to the BRAIN Initiative from which millions of people would benefit.

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7. REFERENCES

1. David Rowitch, Arnold Kriegstein: Developmental genetics of vertebrate glia-cell specification. *Nature* 468, 214-222 (2010) DOI: 10.1038/nature09611
2. World Health Organization: Neurological disorders: public health challenges. *WHO press*, Geneva, Switzerland (2007) No DOI Found
3. Eric Kandel, Henry Markram, Paul Matthews, Rafael Yuste, Christof Koch: Neuroscience thinks big (and collaboratively) *Nat Rev Neurosci* 14, 659-664 (2014) DOI: 10.1038/nrn3578
4. Anna Devor, Peter Bandettini, David Boas, James Bower, Richard Buxton, Lawrence Cohen, Anders Dale, Gaute Einevoll, Peter Fox, Maria Franceschini, Karl Friston, James

Fujimoto, Mark Geyer, Joel Greenberg, Eric Halgren, Matti Hamalainen, Fritjof Helmchen, Bradley Hyman, Alan Jasanoff, Terry Jernigan, Eng Lo, Pierre Magistretti, Joseph Mandeville, Eliezer Masliah, Partha Mitra, William Mobley, Michael Moskowitz, Axel Nimmerjahn, John Reynolds, Bruce Rosen, Brian Salzberg, Chris Schaffer, Gabriel Silva, Peter So, Nicholas Spitzer, Roger Tootell, David Van Essen, Wim Vanduffel, Sergei Vinogradov, Lawrence Wald, Lihong Wang, Bruno Weber, Arjun Yodh: The challenge of connecting the dots in the B.R.A.I.N. *Neuron* 80, 270-274 (2014)
DOI: 10.1016/j.neuron.2013.09.008

5. Lyric Jorgenson, William Newsome, David Anderson, Cornelia Bargmann, Emery Brown, Karl Deisseroth, John Donoghue, Kathy Hudson, Geoffrey Ling, Peter MacLeish, Eve Marder, Richard Normann, Joshua Sanes, Mark Schnitzer, Terrence Sejnowski, David Tank, Roger Tsien, Kamil Ugurbil, John Wingfield: The BRAIN initiative: developing technology to catalyst neuroscience discovery. *Phil Trans R Soc B* 370, 20140164 (2015)
DOI: 10.1098/rstb.2014.0164

6. Mark Schena, Dari Shalon, Ronald Davis, Patrick Brown: Quantitative monitoring of gene expression patterns with a complementary DNA microarray. *Science* 270, 467-470 (1995)
DOI: 10.1126/science.270.5235.467

7. Deval Lashkari, Joseph DeRisi, John McCusker, Allen Namath, Cristl Gentile, Seung Hwang, Patrick Brown Ronald Davis: Yeast microarrays for genome wide parallel genetic and gene expression analysis. *Proc Natl Acad Sci USA* 94, 13057-13062 (1997)
DOI: 10.1073/pnas.94.24.13057

8. Sydney Brenner, Maria Johnson, John Bridgham, George Golda, David Lloyd, Davida Johnson, Shujun Luo, Sarah McCurdy, Michael Foy, Mark Ewan, Rithy Roth, Dave George, Sam Eletr, Glenn Albrecht, Eric Vermaas, Steven Williams, Keith Moon, Timothy Burcham, Michael Pallas, Robert DuBridge, James Kirchner, Karen Fearon, Jen Mao, Kevin Corcoran: Gene expression analysis by massively parallel signature sequencing (MPSS) on microbead arrays. *Nat Biotechnol* 18, 630-634 (2000)
DOI: 10.1038/76469

9. Stephan Schuster: Next-generation sequencing transforms today's biology. *Nat Methods* 5, 16-18 (2008)
DOI: 10.1038/nmeth1156

10. Jose Bras, Rita Guerreiro, John Hardy: Use of next-generation sequencing and other whole-genome strategies to dissect neurological disease. *Nat Rev Neurosci* 13, 453-464 (2012)
DOI: 10.1038/nrn3271

11. Jia-Nee Foo, Jian-Jun Liu, Eng-King Tan: Whole-genome and whole-exome sequencing in neurological diseases. *Nat Rev Neurology* 8, 508-517 (2012)
DOI: 10.1038/nrneurol.2012.148

12. Teng Jiang, Meng-Shan Tan, Lan Tan, Jin-Tai Yu: Application of next-generation sequencing technologies in neurology. *Ann Transl Med* 2, 125 (2014)
No DOI Found

13. Hyo Kang, Yuka Kawasawa, Feng Cheng, Ying Zhu, Xuming Xu, Mingfeng Li, Andre Sousa, Mihovil Pletikos, Kyle Meyer, Goran Sedmak, Tobias Guennel, Yurae Shin, Matthew Johnson, Zeljka Krsnik, Simone Mayer, Sofia Fertuzinhos, Sheila Umlauf, Steven Lisgo, Alexander Vortmeyer, Daniel Weinberger, Shrikant Mane, Thomas Hyde, Anita Huttner, Mark Reimers, Joel Kleinman, Nenad Sestan: Spatio-temporal transcriptome of the human brain. *Nature* 478, 483-489 (2011)
DOI: 10.1038/nature10523

14. Carlo Colantuoni C, Barbara Kipska, Tianzhang Ye, Thomas Hyde, Ran Tao, Jeffrey Leek, Elizabeth Colantuoni, Abdel Elkahloun, Mary Herman, Daniel Weinberger, Joel Kleinman: Temporal dynamics and genetic comtrol of transcriptional in the human prefrontal cortex. *Nature* 478, 519-523 (2011)
DOI: 10.1038/nature10524

15. Michael Hawrylycz, Ed Lein, Angela Guillozet-Bongaarts, Elain Shen, Lydia Ng, Jeremy Miller, Louie van de Lagemaat, Kimberly Smith, Amanda Ebbert, Zackery Riley, Chris Abajian, Christian Beckmann, Amy Bernard, Darren Bertagnolli, Andrew Boe, Preston Cartagena, Mallar Chakravarty, Mike Chapin, Jimmy Chong, Rachel Dalley, Barry Daly, Chinh Dang, Suvro Datta, Nick Dee, Tim Dolbeare, Vance Faber, David Feng, David Fowler, Jeff Goldy, Benjamin Gregor, Zeb Haradon, David Haynor, John Hohmann,

Steve Horvath, Robert Howard, Andreas Jeromin, Jayson Jochim, Marty Kinnunen, Christopher Lau, Evan Lazarz, Changkyu Lee, Tracy Lemon, Ling Li, Yang Li, John Morris, Caroline Overly, Patrick Parker, Sheana Parry, Melissa Redding, Joshua Royall, Jay Schulkin, Pedro Sequeira, Clifford Slaughterbeck, Simon Smith, Andy Sodt, Susan Sunkin, Beryl Swanson, Marquis Vawter, Derric Williams, Paul Wohnoutka, Ronald Zielke, Daniel Geschwind, Patrick Hof, Stephen Smith, Christof Koch, Seth Grant, Allan Jones: An anatomically comprehensive atlas of the adult human brain transcriptome. *Nature* 489, 391-396 (2012)
DOI: 10.1038/nature11405

16. Kazuki Kurimoto, Mitinori Saitou: Single-cell cDNA microarray profiling of complex biological processes of differentiation. *Curr Opin Genet Dev* 20, 470-477 (2010)
DOI: 10.1016/j.gde.2010.06.003

17. Fatih Ozsolak, Alon Goren, Melissa Gymrek, Mitchell Guttman, Aviv Regev, Bradley Bernstein, Patrice Milos: Digital transcriptome profiling from attomole-level RNA samples. *Genome Res* 20, 519-525 (2010)
DOI: 10.1101/gr.102129.109

18. Fatih Ozsolak, David Ting, Ben Wittner, Brian Brannigan, Suchismita Paul, Nabeel Bardeesy, Sridhar Ramaswamy, Patrice Milos, Daniel Haber: Amplification-free digital gene expression profiling from minute cell quantities. *Nat Methods* 7, 619-621 (2010)
DOI: 10.1038/nmeth.1480

19. Tatiana Subkhankulova, Kojiro Yano, Hugh Robinson, Frederick Kivesey: Grouping and classifying electrophysiologically-defined classes of neocortical neurons by single cell, whole-genome expression profiling. *Front. Mol. Neurosci.* 3, 10 (2010)
DOI: 10.3389/fnmol.2010.00010

20. Maria Toledo-Rodriguez, Barak Blumenfeld, Caizhi Wu, Junyi Luo, Bernard Attali, Philip Goodman, Henry Markram: Correlation maps allow neuronal electrical properties to be predicted from single-cell gene expression profiles in rat neocortex. *Cereb. Cortex* 14, 1310-1327 (2004)
DOI: 10.1093/cercor/bhh092

21. Mariela Zirlinger, Gabriel Kreiman, David Anderson: Amygdala-enriched genes identified by microarray technology are restricted to specific amygdaloid subnuclei. *Proc Natl Acad Sci USA* 98, 5270-5275 (2001)
DOI: 10.1073/pnas.091094698

22. Ed Lein, Xinyu Zhao, Fred Gage: Defining a molecular atlas of the hippocampus using DNA microarrays and high-throughput *in situ* hybridization. *J Neurosci* 24, 3879-3889 (2004)
DOI: 10.1523/JNEUROSCI.4710-03.2004

23. Arjun Raj, Alexander van Oudenaarden: Single-molecule approaches to stochastic gene expression. *Annu Rev Biophys* 38, 255-270 (2009)
DOI: 10.1146/annurev.biophys.37.032807.125928

24. Kevin Janes, Chun-Chao Wang, Karin Holmberg, Kristin Cabral, Joan Brugge: Identifying single-cell molecular programs by stochastic profiling. *Nat Methods* 7, 311-317 (2010)
DOI: 10.1038/nmeth.1442

25. Chee Chung, Hyemyung Seo, Kai Sonntag, Andrews Brooks, Ling Lin, Ole Isacson: Cell type-specific gene expression of midbrain dopaminergic neurons reveals molecules involved in their vulnerability and protection. *Hum Mol Genet* 14, 1709-1725 (2005)
DOI: 10.1093/hmg/ddi178

26. Moritz Rossner, Johannes Hirrlinger, Sven Wichert, Christine Boehm, Dieter Newrzella, Holger Hiemisch, Gisela Eisenhardt, Carolin Stuenkel, Oliver von Ahsen, Klaus-Armin Nave: Global transcriptome analysis of genetically identified neurons in the adult cortex. *J Neurosci* 26, 9956-9966 (2006)
DOI: 10.1523/JNEUROSCI.0468-06.2006

27. Charmaine Pietersen, Maribel Lim, Tsung-Ung Woo: Obtaining high quality RNA from single cell populations in human postmortem brain tissue. *J Vis Exp* 30, 1444 (2009)
DOI: 10.3791/1444

28. Paola Arlotta, Bradley Molyneaux, Chen Jinhui, Jun Inoue, Ryo Kominami, Jeffrey Macklis: Neuronal subtype-specific genes that control cortico-spinal motor neuron development *in vivo*. *Neuron* 45, 207-221 (2005)
DOI: 10.1016/j.neuron.2004.12.036

29. Mary Lobo, Stanislav Karsten, Michella Gray, Daniel Geschwind, William Yang: FACS-array profiling of striatal projection neuron subtypes

in juvenile and adult mouse brains. *Nat Neurosci* 9, 443-452 (2006)
DOI: 10.1038/nn1654

30. Eric Marsh, Jennifer Minarcik, Kenneth Campbell, Amy Brooks-Kayal, Jeffrey Golden: FACS-array gene expression analysis during early development of mouse telencephalic interneurons. *Dev Neurobiol* 68, 434-445 (2008)
DOI: 10.1002/dneu.20602

31. Bradley Molyneaux, Paola Arlotta, Ryann Fame, Jessica MacDonald, Kyle MacQuarrie, Jeffrey Macklis: Novel subtype-specific genes identify distinct subpopulations of callosal projection neurons. *J Neurosci* 29, 12343-12354 (2009)
DOI: 10.1523/JNEUROSCI.6108-08.2009

32. John Cahoy, Ben Emery, Amit Kaushal, Lynette Foo, Jennifer Zamanian, Karen Christopherson, Yi Wang, Jane Lubischer, Paul Krieg, Sergey Krupenko, Wesley Thompson, Ben Barres: A transcriptome database for astrocytes, neurons, and oligodendrocytes: a new resource for understanding brain development and function. *J Neurosci* 28, 264-278 (2008)
DOI: 10.1523/JNEUROSCI.4178-07.2008

33. Barbara Barres, Bruce Silverstein, David Corey, Linda Chun: Immunological, morphological, and electrophysiological variation among retinal ganglion cells purified by panning. *Neuron* 1, 791-803 (1988)
DOI: 10.1016/0896-6273(88)90127-4

34. Barbara Barres, Ian Hart, Harriet Coles, Julia Burne, James Voyvodic, William Richardson, Martin Raff: Cell death and control of cell survival in the oligodendrocyte lineage. *Cell* 70, 1-46 (1992)
DOI: 10.1016/0092-8674(92)90531-G

35. Joseph Doyle, Joseph Dougherty, Myriam Heiman, Eric Schmidt, Tanya Stevens, Guojun Ma, Sujata Bupp, Prerana Shrestha, Rajiv Shah, Martin Doughty, Shiaoching Gong, Paul Greengard, Nathaniel Heintz: Application of a translational profiling approach for the comparative analysis of CNS cell types. *Cell* 135, 749-762 (2008)
DOI: 10.1016/j.cell.2008.10.029

36. Myriam Heiman, Anne Schaefer, Shiaoching Gong, Jayms Peterson, Michelle Day, Keri Ramsey, Mayte Suarez-Farinas, Cordelia Schwarz, Dietrich Stephan, James Surmeier, Paul Greengard, Nathaniel Heintz: A translational profiling approach for the molecular characterization of CNS cell types. *Cell* 135, 738-748 (2008)
DOI: 10.1016/j.cell.2008.10.028

37. Elisenda Sanz, Linghai Yang, Thomas Su, David Morris, Stanley McKnight, Paul Amieux: Cell-type-specific isolation of ribosome-associated mRNA from complex tissues. *Proc Natl Acad Sci USA* 106, 13939-13944 (2009)
DOI: 10.1073/pnas.0907143106

38. Joseph Dougherty, Eric Schmidt, Miho Nakajima, Nathaniel Heintz: Analytical approaches to RNA profiling data for the identification of genes enriched in specific cells. *Nucleic Acids Res* 38, 4218-4230 (2010)
DOI: 10.1093/nar/gkq130

39. Michael Forrest: Intracellular calcium dynamics permit a Purkinje neuron model to perform toggle and gain computations upon it inputs. *Front Comput Neurosci* 8, 86 (2014)
DOI: 10.3389/fncom.2014.00086

40. Joshua Shulman, Philip De Jager, Mel Feany: Parkinson's disease: genetics and pathogenesis. *Annu Rev Pathol* 6, 193-222 (2011)
DOI: 10.1146/annurev-pathol-011110-130242

41. Francis Walker: Huntington's disease. *Lancet* 369, 218-228 (2007)
DOI: 10.1016/S0140-6736(07)60111-1

42. Kristjan Jessen, Rhona Mirsky: Glial cells in the enteric nervous system contain glial fibrillary acidic protein. *Nature* 286, 736-737 (1980)
DOI: 10.1038/286736a0

43. Martin Raff, Erika Abney, Jim Cohen, Ronald Lindsay, Mark Noble: Two types of astrocytes in cultures of developing rat white matter: differences in morphology, surface gangliosides, and growth characteristics. *J Neurosci* 3, 1289-1300 (1983)
No DOI Found

44. Voon Yong, Fiona Yong, Andre Olivier, Yves Robitaille, Jack AnTel: Morphologic heterogeneity of human adult astrocytes in culture: correlation with HLA-DR expression. *J Neurosci Res* 27, 678-688 (1990)
DOI: 10.1002/jnr.490270428

45. Mary Bailey, Michael Shipley: Astrocyte subtypes in the rat olfactory bulb:

morphological heterogeneity and differential laminar distribution. *J Comp Neurol* 328, 501-526 (1993)
DOI: 10.1002/cne.903280405

46. Marie Simard, Maiken Nedergaard: The neurobiology of glia in the context of water and ion homeostasis. *Neurosci* 129, 877-896 (2004)
DOI: 10.1016/j.neuroscience.2004.09.053

47. Ralf Dringen, Rolf Gebhardt, Bernd Hamprecht: Glycogen in astrocytes: possible function as lactate supply for neighboring cells. *Brain Res* 623, 208-214 (1993)
DOI: 10.1016/0006-8993(93)91429-V

48. Eric Newman: New roles for astrocytes: regulation of synaptic transmission. *Trends Neurosci* 10, 536-542 (2003)
DOI: 10.1016/S0166-2236(03)00237-6

49. Maiken Nedergaard, Bruce Ransom, Steven Goldman: New roles for astrocytes: redefining the functional architecture of the brain. *Trends Neurosci* 10, 523-530 (2003)
DOI: 10.1016/j.tins.2003.08.008

50. Ben Barres: The mystery and magic of glia: a perspective on their roles in health and disease. *Neuron* 60, 430-440 (2008)
DOI: 10.1016/j.neuron.2008.10.013

51. Takahiro Takano, Guo-Feng Tian, Weiguo Peng, Nanhong Lou, Witold Libionka, Xiaoning Han, Maiken Nedergaard: Astrocyte-mediated control of cerebral blood flow. *Nat Neurosci* 9, 260-267 (2006)
DOI: 10.1038/nn1623

52. Cagla Eroglu, Ben Barres: Regulation of synaptic connectivity by glia. *Nature* 468, 223-231 (2010)
DOI: 10.1038/nature09612

53. Nurit Ballas, Daniel Lioy, Christopher Grunseich, Gail Mandel: Non-cell autonomous influence of MeCP2-deficient glia on neuronal dendritic morphology. *Nat Neurosci* 12, 311-317 (2009)
DOI: 10.1038/nn.2275

54. Shelley Jacobs, Laurie Doering: Astrocytes prevent abnormal neuronal development in the fragile X mouse. *J Neurosci* 30, 4508-4514 (2010)
DOI: 10.1523/JNEUROSCI.5027-09.2010

55. Anna Molofsky, Robert Krenick, Erik Ullian, Hui-hsin Tsai, Benjamin Deneen, William Richardson, Ben Barres, David Rowitch: Astrocytes and disease: a neurodevelopmental perspective. *Genes Dev* 26, 891-907 (2012)
DOI: 10.1101/gad.188326.112

56. Alexei Verkhratsky, Michael Sofroniew, Albee Messing, Nihal deLanerolle, David Rempe, Jose Rodriguez, Maiken Nedergaard: Neurological diseases as primary gliopathies: a reassessment of neurocentrism. *ASN Neuro* 4, e00082 (2012)
DOI: 10.1042/AN20120010

57. Dorte Pelvig, Henning Pakkenberg, Anette Stark, Bente Pakkenberg: Neocortical glial cell numbers in human brains. *Neurobiol Aging* 29, 1754-1762 (2008)
DOI: 10.1016/j.neurobiolaging.2007.04.013

58. Goichi Miyoshi, Gord Fishell: Directing neuron-specific transgene expression in the mouse CNS. *Curr Opin Neurobiol* 16, 577-584 (2006)
DOI: 10.1016/j.conb.2006.08.013

59. Sandra Kuhlman, Josh Huang: High-resolution labeling and functional manipulation of specific neuron types in mouse brain by Cre-activated viral gene expression. *PLoS One* 3, e2005 (2008)
No DOI Found

60. Benjamin Okaty, Ken Sugino, Sacha Nelson: Cell type-specific transcriptomics in the brain. *J. Neurosci* 31, 6939-6943 (2011)
DOI: 10.1523/JNEUROSCI.0626-11.2011

61. Benjamin Okaty, Ken Sugino, Sacha Nelson: A quantitative comparison of cell-type-specific microarray gene expression profiling methods in the mouse brain. *PLoS One* 6, e16493 (2011)
DOI: 10.1371/journal.pone.0016493

62. Jason Dugas, Yu-chuan Tai, Terence Speed, John Ngai, Ben Barres: Functional genomic analysis of oligodendrocyte differentiation. *J. Neurosci* 26, 10967-10983 (2006)
DOI: 10.1523/JNEUROSCI.2572-06.2006

63. Ditte Lovatt, Ursula Sonnewald, Helle Waagepetersen, Arne Schousboe, Wei He, Jane Lin, Xiaoning Han, Takahiro Takano, Su Wang, Fraser Sim, Steven Goldman, Maiken Nedergaard: The transcriptome and metabolic gene signature of protoplasmic astrocytes in the adult murine cortex. *J Neurosci* 27, 12255-12266 (2007)
DOI: 10.1523/JNEUROSCI.3404-07.2007

64. Pierre Lau, Jonathan Verrier, Joseph Nielsen, Kory Johnson, Lucia Notterpek, Lynn Hudson: Identification of dynamically regulated microRNA and mRNA networks in developing oligodendrocytes. *J Neurosci* 28, 11720-11730 (2008)
DOI: 10.1523/JNEUROSCI.1932-08.2008

65. Richard Daneman, Lu Zhou, Dritan Agalliu, John Cahoy, Amit Kaushal, Ben Barres: The mouse blood-brain barrier transcriptome: a new resource for understanding the development and function of brain endothelial cells. *PLoS One* 5(10), e13741 (2010)
DOI: 10.1371/journal.pone.0013741

66. Christi Wylie, Timothy Hendricks, Bing Zhang, Lily Wang, Pengcheng Lu, Patrick Leahy, Stephanie Fox, Hiroshi Maeno, Evan Deneris: Distinct transcriptomes define rostral and caudal serotonin neurons. *J Neurosci* 30, 670-684 (2010)
DOI: 10.1523/JNEUROSCI.4656-09.2010

67. Bernd Friedrich, Philipp Euler, Ruhtraut Ziegler, Alexandre Kuhn, Bernhard Landwehrmeyer, Ruth Luthi-Carter, Cornelius Weiller, Sabine Hellwig, Birgit Zucker: Comparative analyses of Purkinje cell gene expression profiles reveal shared molecular abnormalities in models of different polyglutamine diseases. *Brain Res* 1481, 37-48 (2012)
DOI: 10.1016/j.brainres.2012.08.005

68. Oliver Bracko, Tatjana Singer, Stefan Aigner, Marlen Knobloch, Beate Winner, Jasodhara Ray, Gregory Clemenson Jr, Hoonkyo Suh, Sebastien Couillard-Despres, Ludwig Aigner, Fred Gage, Sebastian Jessberger: Gene expression profiling of neural stem cells and their neuronal progeny reveals IGF2 as a regulator of adult hippocampal neurogenesis. *J Neurosci* 32, 3376-3387 (2012)
DOI: 10.1523/JNEUROSCI.4248-11.2012

69. Clara Beutner, Bettina Linnartz-Gerlach, Susanne Schmidt, Marc Beyer, Michael Mallmann, Andrea Staratschek-Jox, Joachim Schultze, Harald Neumann: Unique transcriptome signature of mouse microglia. *Glia* 61, 1429-1442 (2013)
DOI: 10.1002/glia.22524

70. Marie Orre, Willem Kamphuis, Lana Osborn, Jeroen Melief, Lieneke Kooijman, Inge Huitinga, Jan Klooster, Koen Bossers, Elly Hol: Acute isolation and transcriptome characterization of cortical astrocytes and microglia from young and aged mice. *Neurobiol Aging* 35, 1-14 (2014)
DOI: 10.1016/j.neurobiolaging.2013.07.008

71. Isaac Chiu, Emiko Morimoto, Hani Goodarzi, Jennifer Liao, Sean O'Keeffe, Hemali Phatnani, Michael Muratet, Michael Carroll, Shawn Levy, Saeed Tavazoie, Richard Myers, Tom Maniatis: A neurodegeneration-specific gene-expression signature of acutely isolated microglia from an amyotrophic lateral sclerosis mouse model. *Cell Rep* 4, 384-401 (2013)
DOI: 10.1016/j.celrep.2013.06.018

72. Urmi Bandyopadhyay, Justin Cotney, Maria Nagy, Sunghee Oh, Jing Leng, Milind Mahajan, Shrikant Mane, Wayne Fenton, James Noonan, Arthur Horwich: RNA-seq profiling of spinal cord motor neurons from a presymptomatic SOD1 ALS mouse. *PLoS One* 8, e53575 (2013)
DOI: 10.1371/journal.pone.0053575

73. Ye Zhang, Kenian Chen, Steven Sloan, Mariko Bennett, Anja Scholze, Sean O'Keeffe, Hemali Phatnani, Paolo Guarnieri, Christine Caneda, Nadine Ruderisch, Shuyun Deng, Shane Liddelow, Chaolin Zhang, Richard Daneman, Tom Maniatis, Ben Barres, Jai-qian Wu: An RNA-sequencing transcriptome and splicing database of glia, neurons, and vascular cells of the cerebral cortex. *J Neurosci* 34, 11929-11947 (2014)
DOI: 10.1523/JNEUROSCI.1860-14.2014

74. Jessica Lerch, Frank Kuo, Dario Motti, Richard Morris, John Bixby, Vance Lemmon: Isoform diversity and regulation in peripheral and central neurons revealed through RNA-seq. *PLoS One* 7, e30417 (2012)
DOI: 10.1371/journal.pone.0030417

75. Eric Lai, Bergin Tam, Gerald Rubin: Pervasive regulation of *Drosophila* Notch target genes by GY-box-, Brd-box-, and K-box-class microRNAs. *Genes Dev* 19, 1067-1080 (2005)
DOI: 10.1101/gad.1291905

76. Fiona Doetsch, Isabelle Caille, Daniel Lim, Jose Garcia-Verdugo, Arturo Alvarez-Buylla: Subventricular zone astrocytes are neural stem cells in the adult mammalian brain. *Cell* 97, 703-716 (1999)
DOI: 10.1016/S0092-8674(00)80783-7

77. Linda Edman, Helena Mira, Ernest Arenas: The beta-chemokines CCL2 and CCL7 are

two novel differentiation factors for midbrain dopaminergic precursors and neurons. *Exp Cell Res* 314, 2123-2130 (2008)
DOI: 10.1016/j.yexcr.2008.02.019

78. eMalick Njie, Ellen Boelen, Frank Stassen, Harry Steinbusch, David Borchelt, Wolfgang Streit: *Ex vivo* cultures of microglia from young and aged rodent brain reveal age-related changes in microglial function. *Neurobiol. Aging* 33, 195.e1-195.e12 (2012)
DOI: 10.1016/j.neurobiolaging.2010.05.008

79. Amanda Sierra, Andres Gottfried-Blackmore, Bruce McEwen, Karen Mulloch: Microglia derived from aging mice exhibit an altered inflammatory profile. *Glia* 55, 412-424 (2007)
DOI: 10.1002/glia.20468

80. Ali Mortazavi, Brian Williams, Kenneth McCue, Lorian Shaeffer L, Barbara Wold: Mapping and quantifying mammalian transcriptomes by RNA-seq. *Nat Methods* 5, 621-628 (2008)
DOI: 10.1038/nmeth.1226

81. Fuchou Tang, Catalin Barbacioru, Ellen Nordman, Nanlan Xu, Vladimir Bashkirov, Kaiqin Lao, Azim Surani: RNA-seq analysis to capture the transcriptome landscape of a single cell. *Nat Protoc* 5, 516-535 (2010)
DOI: 10.1038/nprot.2009.236

82. Mireille Belanger, Igor Allaman, Pierre Magistretti: Brain energy metabolism: focus on astrocyte-neuron metabolic cooperation. *Cell Metab* 14, 724-738 (2011)
DOI: 10.1016/j.cmet.2011.08.016

83. Anne-Karine Bouzier-Sore, Luc Pellerin: Unraveling the complex metabolic nature of astrocytes. *Front Cell Neurosci* 7, 179 (2013)
DOI: 10.3389/fncel.2013.00179

84. Vincent Lobon, Kitt Petersen, Gary Cline, Jun Shen, Graeme Mason, Sylvie Dufour, Kevin Behar, Gerald Shulman: Astroglial contribution to brain energy metabolism in humans revealed by ^{13}C nuclear magnetic resonance spectroscopy: elucidation of the dominant pathway for neurotransmitter glutamate repletion and measurement of astrocytic oxidative metabolism. *J. Neurosci.* 22, 1523-1531 (2002)
No DOI Found

85. Yoshiaki Itoh, Takanori Esaki, Kazuaki Shimoji, Michelle Cook, Mona Law, Elaine Kaufman, Louis Sokoloff: Dichloroacetate effects on glucose and lactate oxidation by neurons and astroglia *in vitro* and on glucose utilization by brain *in vivo*. *Proc Natl Acad Sci USA* 100, 4879-4884 (2003)
DOI: 10.1073/pnas.0831078100

86. Wolfgang Walz, Srimathie Mukerji: Lactate release from cultured astrocytes and neurons: a comparison. *Glia* 1, 366-370 (1988)
DOI: 10.1002/glia.440010603

87. Ken McCarthy, Jean de Vellis: Preparation of separate astroglial and oligodendroglial cell cultures from rat cerebral tissue. *J Cell Biol* 85, 890-902 (1980)
DOI: 10.1083/jcb.85.3.890

88. Marie Simard, Gregory Arcuino, Takahiro Takano, Qing Liu, Maiken Nedergaard: Signaling at the gliovascular interface. *J Neurosci* 23, 9254-9262 (2003)
No DOI Found

89. Andrea Volterra, Jacopo Meldolesi: Astrocytes, from brain glue to communication elements: the revolution continues. *Nat Rev Neurosci* 6, 626-640 (2005)
DOI: 10.1038/nrn1722

90. Alexander Wilhelm, Walter Volknandt, David Langer, Christine Nolte, Helmut Kettenmann, Herbert Zimmermann: Localization of SNARE proteins and secretory organelle proteins in astrocytes in vitro and in situ. *Neurosci Res* 48. 249-257 (2004)
DOI: 10.1016/j.neures.2003.11.002

91. Jack Wang, Noelia Kunzevitzky, Jason Dugas, Meghan Cameron, Ben Barres, Jeffrey Goldberg: Disease gene candidates revealed by expression profiling of retinal ganglion cell development. *J Neurosci* 27, 8593-8603 (2007)
DOI: 10.1523/JNEUROSCI.4488-06.2007

92. Peter LoVerso, Feng Cui: Cross-species transcriptomic comparison of in vitro and in vivo mammalian neural cells. *Bioinfo Biol Insights*, in press (2015)
No DOI Found

Abbreviations: LCM: laser-capture microdissection; LDM: laser-directed microdissection; FACS: fluorescence-activated cell sorting; PAN: immunopanning; TRAP: translating ribosome affinity purification; NGS: next-generation sequencing

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