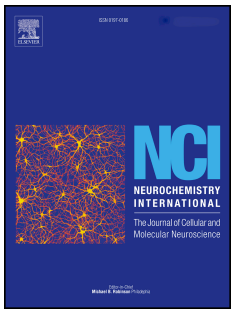
Accepted Manuscript



The neuropharmacology of butyrate: The bread and butter of the microbiota-gut-brain axis?

Roman M. Stilling, Marcel van de Wouw, Gerard Clarke, Catherine Stanton, Timothy G. Dinan TG, John F. Cryan

PII: S0197-0186(16)30174-7

DOI: 10.1016/j.neuint.2016.06.011

Reference: NCI 3889

To appear in: *Neurochemistry International*

Received Date: 25 April 2016

Revised Date: 30 May 2016

Accepted Date: 21 June 2016

Please cite this article as: Stilling, R.M., van de Wouw, M., Clarke, G., Stanton, C., Dinan TG, T.G., Cryan, J.F., The neuropharmacology of butyrate: The bread and butter of the microbiota-gut-brain axis?, *Neurochemistry International* (2016), doi: 10.1016/j.neuint.2016.06.011.

The Neuropharmacology of Butyrate:
The Bread and Butter of the Microbiota-Gut-Brain Axis?

Roman M. Stilling[#,1,2], Marcel van de Wouw[1,2], Gerard Clarke[1,3], Catherine Stanton[1,4], Timothy G. Dinan TG[1,3], John F. Cryan[#1,2]

[1] APC Microbiome Institute, University College Cork, Cork, Ireland
[2] Department of Anatomy and Neuroscience, University College Cork, Cork Ireland
[3] Department of Psychiatry and Neurobehavioural Science, University College Cork, Cork, Ireland
[4] Food Biosciences Department, Teagasc Food Research Centre , Moorepark, Fermoy , Co. Cork , Ireland

Invited review for Neurochemistry International

Correspondence to:
Roman M Stilling, PhD
APC Microbiome Institute /
Dept. Anatomy and Neuroscience
University College Cork
Western, Rd, Cork
Ireland
Tel.: +353(0)21-490-1712
E-Mail: roman.stilling@gmail.com

or

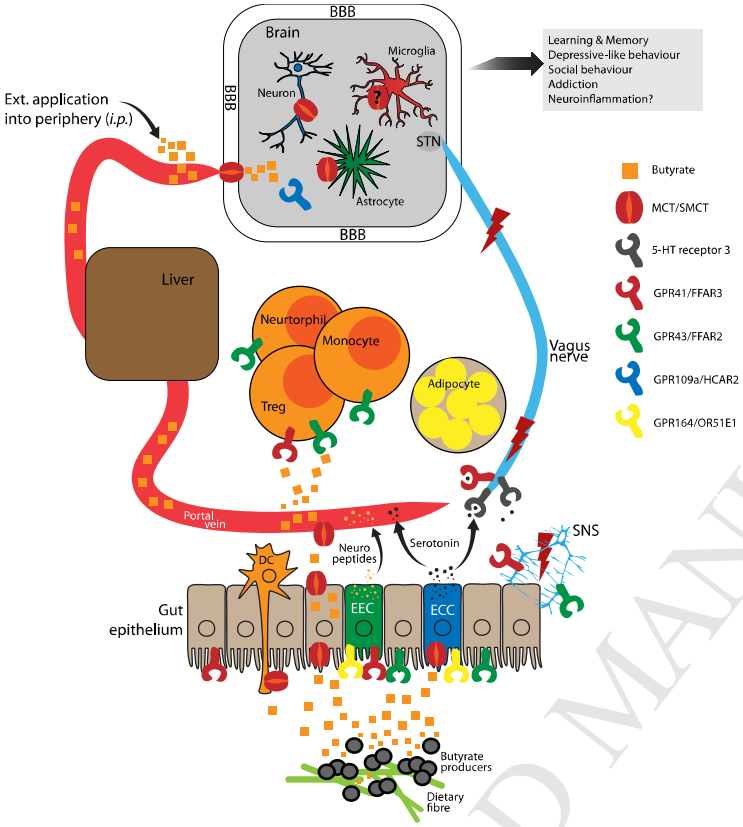
John F. Cryan, Ph.D.
Professor & Chair, Dept Anatomy & Neuroscience
Principal Investigator APC Microbiome Institute
University College Cork,
Room 386, Western Gateway Building,
Western Rd., Cork
Ireland
Tel: +353 21 420 5426
Fax:+353 21 427 3518
E-Mail: j.cryan@ucc.ie

1 Abstract

Several lines of evidence suggest that brain function and behaviour are influenced by microbial metabolites. Key products of the microbiota are short-chain fatty acids (SCFAs), including butyric acid. Butyrate is a functionally versatile molecule that is produced in the mammalian gut by fermentation of dietary fibre and is enriched in butter and other dairy products. Butyrate along with other fermentation-derived SCFAs (e.g. acetate, propionate) and the structurally related ketone bodies (e.g. acetoacetate and D-β-hydroxybutyrate) show interesting effects in various diseases including obesity, diabetes, inflammatory (bowel) diseases, and colorectal cancer as well as neurological disorders. Indeed, it is clear that host energy metabolism and immune functions critically depend on butyrate as a potent regulator, highlighting butyrate as a key mediator of host-microbe crosstalk. In addition to specific receptors (GPR43/FFAR2; GPR41/FFAR3; GPR109a/HCAR2) and transporters (MCT1/SLC16A1; SMCT1/SLC5A8), its effects are mediated by utilisation as an energy source via the β-oxidation pathway and as an inhibitor of histone deacetylases (HDACs), promoting histone acetylation and stimulation of gene expression in host cells. The latter has also led to the use of supraphysiological doses of butyrate as an experimental drug in models for neurological disorders ranging from depression to neurodegenerative diseases and cognitive impairment.

Here we provide a critical review of the literature on butyrate and its effects on multiple aspects of host physiology with a focus on brain function and behaviour. We find fundamental differences in natural butyrate at physiological concentrations and its use as a neuropharmacological agent at rather high, supraphysiological doses in brain research. Finally, we hypothesize that butyrate and other volatile SCFAs produced by microbes may be involved in regulating the impact of the microbiome on behaviour including social communication.

2 Graphical Abstract



3 Highlights

- Butyrate is produced by specific bacteria, mainly in the colon, and is taken up by the host
- Butyrate affects multiple host physiological processes via specific transporters/receptors and as an HDAC inhibitor
- Supraphysiological doses of butyrate exert potent neuropharmacological effects, facilitating synaptic tagging and capturing
- Physiological levels of butyrate may influence brain function indirectly via regulating immune responses and vagus nerve stimulation
- Microbiota-derived volatile butyrate may be involved in host behaviour including social communication

4 Keywords:

neuroepigenetics; sociability; endocrine; Treg; gut-brain axis; nutrition

5 Introduction

The gastrointestinal tract is the main interface for interaction and nutrient exchange between an animal's interior milieu and the outside world. This interface is colonized by a vast and complex microbial ecosystem, which symbiotically interacts with the host. During the last decade, evidence has rapidly accumulated, showing that this microbiota has extensive regulatory effects on host physiology and function of virtually all organ systems (Clarke et al., 2014). As such, central nervous system function and subsequently also human and animal behaviour is influenced by microbial presence, metabolism and activity (Collins et al., 2012; Cryan and Dinan, 2012; Mayer et al., 2014; Sampson and Mazmanian, 2015). The microbiota-gut-brain axis integrates various routes of communication, including endocrine, vagus nerve-dependent and immune signalling as well as direct action of microbial metabolites as signalling molecules in the brain (Clarke et al., 2014; El Aidy et al., 2014; Forsythe et al., 2014; Lyte, 2013; Selkrig et al., 2014; Stilling et al., 2014b). Among the most important and pleiotropic functional components of microbe-to-host signalling are short-chain fatty acids (SCFAs), small organic monocarboxylic acids with less than six carbon atoms, that are major microbial metabolites produced during anaerobic fermentation in the gut (Roy et al., 2006).

The C4 monocarboxylic acid butyric acid (IUPAC name: butanoic acid) is a SCFA that got its name from the Greek word for butter and is infamous for its strong smell of rancid milk or butter, where it is generated from butyric acid-containing triglycerides present in milk fat by lipase-catalysed hydrolysis (Reineccius and Heath, 2006). Contributing to the characteristics of body odour, it is also largely responsible for the smell of vomit and sweat, where it is produced from lipids (e.g. milk fat in the stomach or sebum secreted by sebaceous glands on the skin) by salivary or gastric lipases or bacteria-derived lipases (e.g. by members of *Corynebacterium, Staphylococcus* and *Micrococcus* genera*)* (Holt, 1971).

Butyric acid comes in two isoforms, known as n-butyric acid and iso-butyric acid (**Fig. 1A**). Since n-butyric acid concentrations are outnumbering iso-butyric acid concentrations approximately 5-to-8-fold in human faeces (Payne et al., 2011; Siigur et al., 1993), and only n-butyrate has some of the molecular/pharmacological characteristics discussed in this review, we will focus predominantly on n-butyric acid. We will further refer to is as butyrate as in solution with a pH > pK_a(=4.82), butyric acid appears mainly in its deprotonated form (e.g. in blood of pH 7.4 almost all butyric acid dissociates to butyrate and H^+ (ratio [A-]:[HA] = 380:1)). In the human colon, butyric acid contributes to the slight acidity with a typical pH of about 5.7 to 6.7 ([A-]:[HA] ratios approximately 7.6:1 to 76:1) (Fallingborg, 1999).

Butyrate, the anionic part of dissociated butyric acid and its salts, has been implicated in various host physiological functions including energy homeostasis, obesity, immune system regulation, cancer, and even brain function (Bourassa et al., 2016; Di Sabatino et al., 2005; Li, 2014). Yet, the molecular mechanisms mediating these functions may differ, ranging from metabolic effects to receptor signalling and enzymatic inhibition, and are not completely understood (Canani et al., 2011). Under physiological conditions, i.e. butyrate is only derived from fermentation of dietary fibre in the gut and reaches the circulation in variable µ-molar concentrations, butyrate mainly affects intestinal and adjacent tissues in a significant and mostly beneficial manner ((Canani et al., 2011; Hamer et al., 2008), see sections 7.1, 7.2 and 9). However, butyrate is also widely used as an experimental pharmacological tool compound, and more recently also in neuroscience research, often administered systemically at concentrations of 100-1200mg/kg (Bourassa et al., 2016; Fischer et al., 2010). It is thus of particular interest to the field of microbiota-gut-brain axis research to understand how gut-derived butyrate influences brain function and behaviour.

In this review, we will summarize what is known about the biological relevance of butyrate with a focus on the gut microbiota as its prime source and the known and potential effects butyrate has on brain function and behaviour.

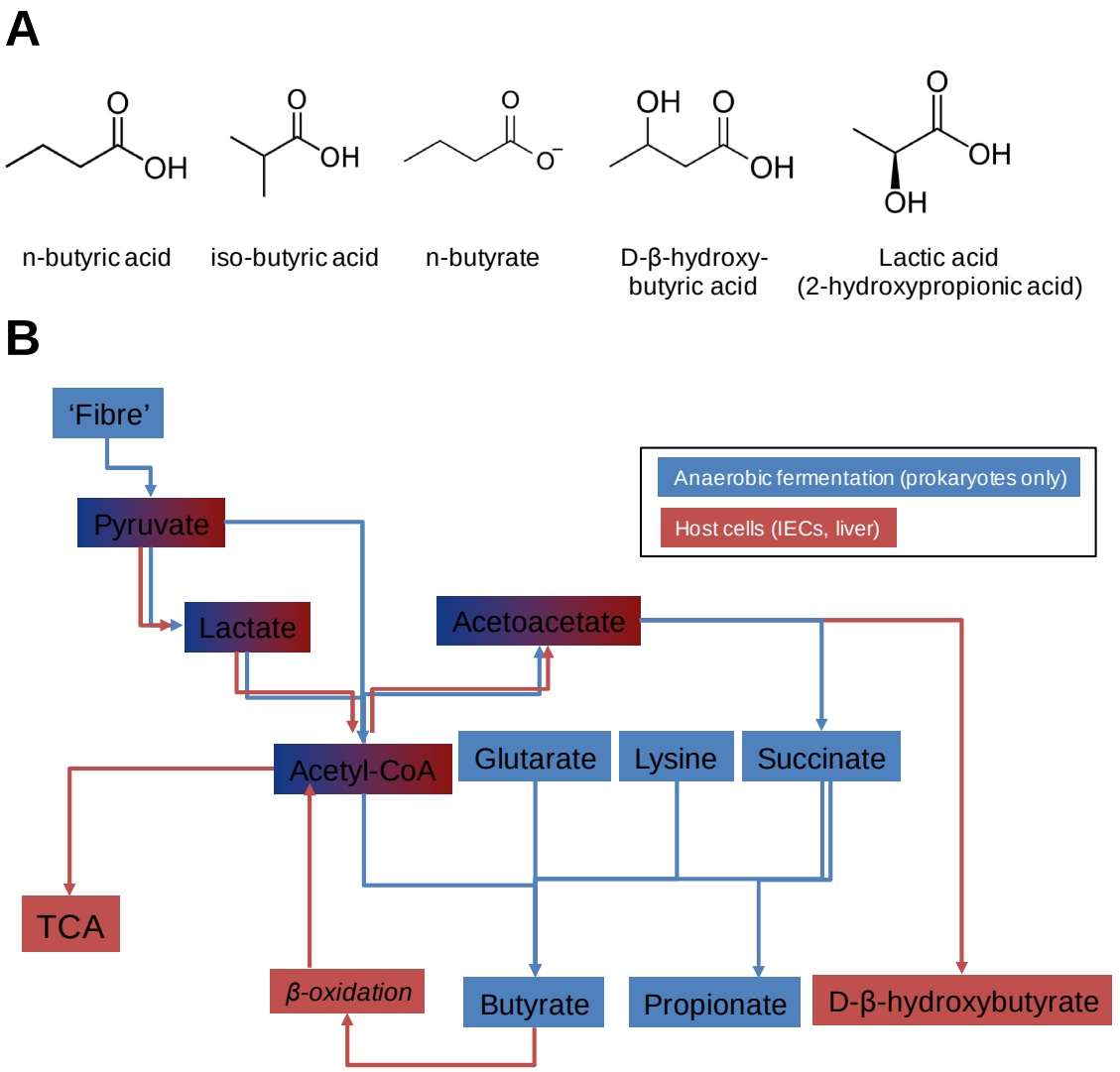


Fig. 1: (A) *Structual representations of butyrate and related molecules.* ***(B)*** *Over-simplified diagram of host-microbiota co-metabolism of butyrate. For more details see (Louis and Flint, 2009; Macfarlane and Macfarlane, 2003). Acetyl-CoA: acetyl coenzyme A; TCA: Tricarboxylic acid cycle (citric acid cycle/Krebs cycle)*

6 Biochemistry

Caecal and colonic fermentation of dietary fibre, carbohydrates and proteins are complex energy-releasing processes that occur under anaerobic conditions and are necessary for survival of many gut-colonising bacterial and fungal species. The main end-products of the different fermentation processes are the SCFAs acetate (C2), propionate (C3) and butyrate (C4), but also - to a lesser extent - so-called branched short-chain fatty acids (iso-butyrate, valerate and iso-valerate) (Fernandes et al., 2014)

In nature, the majority of butyrate is synthesised during anaerobic microbial fermentation of polysaccharides, indigestible to the host, who cannot produce any butyrate on its own. As an exception, butyrate can also be produced by host lipases from the triglyceride tributyrin, used as a prodrug to deliver biologically active butyrate (Gaschott et al., 2001; Miyoshi et al., 2011). Among the microbiota, several synthetic pathways have been described. Apart from the most prevalent Acetyl-Coenzyme A (AcCoA) pathway, where AcCoA is produced via pyruvate derived from fermentation of host-indigestible carbohydrates (known as 'fibre', **Box 1**) or via lactate (Duncan et al., 2004b), there are three alternative synthesis pathways described, starting from glutarate, lysine, and succinate, which converge on the intermediate product crotonyl-CoA (Vital et al., 2014) (**Fig. 1B**). The final step in butyrogenesis is the conversion of either butyryl-phosphate to butyrate by the butyrate kinase (EC 2.7.2.7), encoded by the *buk* gene, or butyry-CoA to butyrate by the butyryl-CoA:acetate CoA-transferase, encoded by the *but* gene. The latter appears to be the main pathway used by butyrate-producing bacteria in the gut (Louis et al., 2004). In contrast, iso-butyrate is mainly produced by fermentation of certain amino acids, i.e. polypeptides and proteins (Zarling and Ruchim, 1987). Butyrate can be further utilised by host cells where it is metabolised in the mitochondrial β-oxidation process that generates NADH, H^+ and AcCoA, which in turn can further be used to generate ATP in the citric acid cycle in the mitochondria (Astbury and Corfe, 2012). In fact, in the host, butyrate is both taken up and metabolised for energy rather quickly, given that even high doses of oral or intravenous butyrate result in relatively rapid peak plasma concentrations (less an hour) and quickly decay within a few hours (Egorin et al., 1999; Kim et al., 2013). On the other hand, the prodrug tributyrin is used to release butyrate at a slower rate (Egorin et al., 1999; Miyoshi et al., 2011).

Box 1: Dietary fibre is a rather unspecific term comprising all host-indigestible dietary carbohydrates, i.e. polysaccharides mainly found in plants and mammalian milk and dairy products (Topping and Clifton, 2001). They can be further subdivided by their solubility in water, their specific sugar monomer and/or polymerisation complexity. An important, well-studied class of soluble fibre is short (3 to 10 monomers) oligosaccharides made from fructose or galactose (FOS and GOS). These can be found for example in agave, bananas, onions, garlic and Jerusalem artichoke. GOS is also found in (breast)milk. Other, glucose-based fibre classes are resistant starch (RS), found in e.g. cooled boiled potatoes, β-glucans, found in oat, barley, wheat, and rye, and cellulose, the main plant cell wall component. In addition, non-starch polymers of xylose and other sugars (xylans and other hemicelluloses) as well as uronic acids (pectins) and cellulose are found in plant-based diets, most prominently in pears, apples, guavas, plums, and oranges (Bindelle et al., 2008). Prebiotics are defined as food supplements that specifically promote growth of health-associated bacteria in the gut. They also are usually non-digestible carbohydrates that reach the caecum to become substrates for microbial fermentation (Cummings et al., 2001; Topping and Clifton, 2001).

A structurally related compound, though with very different origin, is the ketone body D-β-hydroxybutyrate (DHB), which can be synthesized by host cells from AcCoA under conditions like fasting, a ketogenic diet, type I diabetes, or alcoholic ketoacidosis. The major source for DHB is the liver, but there is also some evidence indicating that astrocytes are capable of producing DHB and shuttling it to neurons (Guzmán and Blázquez, 2004), where it can bind fatty acid receptors (see 7.4.2), be metabolised for energy or serve as substrate for amino-acid neurotransmitter synthesis (Yudkoff et al., 2001). Interestingly butyrate and DHB appear to metabolically interact: High levels of butyrate and a ketogenic diet, consisting of mainly lipids and very low amounts of carbohydrates, can increase levels of DHB in blood and in the cerebrospinal fluid (CSF) in calves (Iriki et al., 2009) and the authors suggest that increased DHB is at least partially derived from the additionally supplied butyrate. Due to their structural relatedness, DHB has additional molecular features that mimic those of butyrate and will therefore be discussed along with butyrate throughout this review.

6.1 Sources

6.1.1 Diet

Butyrate—occurs in dairy products in considerable amounts, e.g. whole cow's milk (~0.1g/100g), butter (~3g/100g), cheese (especially goat's cheese (~1-1.8g/100g) and

parmesan (~1.5g/100g; data retrieved from the USDA National Nutrient Database for Standard Reference, Release 28), where it is present due to microbial anaerobic fermentation of fibre, including cellulose, in the ruminant gut. It is also present in human breast milk, resulting in uptake of an estimated amount of approximately 30mg/kg in a breast-fed baby (Aitoro et al., 2015).

6.1.2 The Microbiota

Many bacterial species colonising the colon are capable of digesting (fermenting) fibre in the absence of oxygen. Interestingly, these species do not constitute a monophyletic group, i.e. not all butyrate-producing bacteria are closely related, indicating that the ability for butyrate production must have occurred repeatedly during host-microbe co-evolution. Most of the butyrogenic bacteria belong to the Firmicutes phylum, within which the Clostridium clusters IV and XIVa, 16S-rRNA-defined phylogenetic groups belonging to the class Clostridia, are the best-studied groups (Barcenilla et al., 2000; Collins et al., 1994; Kläring et al., 2013; Pryde et al., 2002; Stackebrandt et al., 1999). For a detailed list of selected butyrate-producing bacteria see (Li and Li, 2014).

In addition to these, microbiota composition as a whole can have an effect on overall production efficiency of butyrate and other SCFAs. As such, acetate and lactate formed during fermentation are important contributors to butyrate production, hence a significant amount of the butyrate made is not directly derived from fibre by particular species, but by interactions within the gut microbial ecosystem (Duncan et al., 2004a, 2004b; Flint et al., 2007; Veiga et al., 2014). Intriguingly, complex inter-species cross-feeding mechanisms have been described that link the metabolic activity of lactic acid bacteria (e.g., lactobacilli and bifidobacteria, key species generally considered to be probiotic, i.e. to promote host health) with butyrate-producing pathways (Belenguer et al., 2006; De Vuyst and Leroy, 2011; Flint et al., 2007; Rios-Covian et al., 2015; Rivière et al., 2015). In this context it is interesting to note that in extreme cases such as the short-bowel syndrome in humans, fermentation-derived overdoses of lactate (2-3mM in serum) can induce severe neurological symptoms (Ewaschuk et al., 2005), which might be treated by enhancing conversion of lactate to butyrate in the colonic lumen. In addition, luminal pH and SCFA concentrations have been found to be inversely related (Cummings et al., 1987) which is in line with butyrogenic bacteria being able to proliferate at a lower pH as compared to gram negative Bacteroides species, more prevalent at a higher pH (Duncan et al., 2009; Walker et al., 2005). Therefore, also the intracolonic milieu significantly affects butyrate production efficiency by the microbiota.

Finally, butyrate can also be produced from mucin degradation. Mucins are heavily glycosylated proteins expressed by the host epithelium forming the basis of the mucus lining

of the intestinal tract. This particular form of symbiosis whereby the host actively contributes to colonisation of specific mucin-associated bacteria is of particular importance since this in itself is regulated by specific mucin expression and glycosylation patterns. Mucin-degrading butyrate-producing bacteria from several Clostridial clusters, including species *Rosburia intestinalis* and *Eubacterium rectale,* have just recently been identified since they are tightly associated with epithelial mucus (Van den Abbeele et al., 2013) and may thus not be represented appropriately in faecal samples. A recent study found that 3 out of 7 mucin-feeding bacteria were able to produce butyrate (Levine et al., 2013) and it is likely that more novel butyrogenic species will be found in human intestines in the near future.

While butyrate from the diet is readily available to host tissues, presumably release of butyrate from fermentation is slower and more steady over time, likely resulting in a more complex profile of baseline and peak concentrations under day-to-day conditions in a human non-experimental situation.

7 Concentrations and Transport – Butyrate regulates host physiology

7.1 Intestinal Synthesis and Concentrations – Relevance to Host Metabolism and Obesity

As butyrate is – with few exceptions in tissues of goats, rabbits and piglets (Kien et al., 2000; Nandedkar et al., 1969; Nandedkar and Kumar, 1969) – almost exclusively produced by gut bacteria, or taken up with the diet, butyrate concentrations are highest in the gut lumen. Human faeces show substantial variability in faecal butyrate concentrations (McOrist et al., 2011) in the range of about 3.5 to 32.6 g/kg of butyrate, as well as ~60 g/kg acetate and ~10-20 g/kg propionate (Macfarlane and Macfarlane, 2003; McOrist et al., 2011). This is in line with the typically cited ratio of about 60:20:20 for acetate, propionate and butyrate in colon and stool (den Besten et al., 2013). Notably, butyrate is the main source for energy metabolism in intestinal epithelial cells, especially colonocytes (den Besten et al., 2013), so that a significant proportion of microbial-released butyrate is rapidly taken up and consumed locally in the gut (Hamer et al., 2008; Topping and Clifton, 2001), which also means that faecal concentrations do not necessarily represent SCFA production rates or concentrations present in the colon (den Besten et al., 2013; Rechkemmer et al., 1988; Verbeke et al., 2015). Hence, it is important to note that variations in faecal SCFA concentrations could be a result of either, altered production or altered colonic absorption. This host-microbe co-metabolism is of particular importance for studies comparing faecal microbiota composition in case/control studies or when interpreting correlations between faecal SCFA content with a certain physiological parameter. In short, a highly fermenting gut promotes caloric extraction

from the diet, which may have been an evolutionary advantage at times of limited food resources but results in quicker weight gain in individuals carrying a high number of these bacteria, despite the otherwise positive effects of this fermentation as outlined above. It thus remains to be elucidated how body-mass-index, fibre intake, microbiota composition and faecal SCFA concentrations are correlated (Clarke et al., 2014; Fernandes et al., 2014). This will be important to define to inform microbiota-directed interventions that might inadvertently impact on butyrate production.

In addition, butyrate and other SCFAs have more remote, indirect effects on host metabolism. It is now well established that SCFAs modulate colonic motility by stimulating serotonin secretion from gut enterochromaffin cells (Fukumoto et al., 2003; Reigstad et al., 2015; Yano et al., 2015), which are known as signalling interfaces between gut microbiota and host (Rhee et al., 2009), at least in part through activation of the vagus nerve via the 5-HT_3 receptor (Fukumoto et al., 2003).

7.2 Transport, circulation and turnover in the host

It is important to note that butyrate can also cross the epithelial barrier and enter the circulation via the hepatic portal vein, which connects the gastrointestinal tract, spleen and liver (Peters et al., 1992). While concentrations in the portal vein are still considerable (~18µmol/l in a fasting human, 14-64µmol/l in sudden death victims (Cummings et al., 1987; Hamer et al., 2008; Peters et al., 1992), concentrations in peripheral blood appear to be relatively low (lower µ-molar range, ~20% of portal vein concentrations) (Cummings et al., 1987; Jakobsdottir et al., 2013; Peters et al., 1992). However, because butyrate is rapidly metabolised in the periphery, short-term peak concentrations might be much higher. In contrast, acetate and propionate are detectable in peripheral blood at about 50% and 10%, respectively, of concentrations found in hepatic portal blood. Therefore, the liver appears to represent a major sink for gut-produced SCFAs, where they may be metabolized via β-oxidation, used for the synthesis of ketone bodies or converted to AcCoA (Bach Knudsen et al., 2003; Cummings et al., 1987), while high systemic acetate levels likely represents production by the host in a large variety of cells from AcCoA by acetyl-CoA hydrolase (Knowles et al., 1974; Peters et al., 1992). However, more recent studies found that peripheral blood SCFA levels correlate with dietary intake of fibre, suggesting butyrate is transported through the circulation and that other organs may be affected by changes in butyrate concentration (Tarini and Wolever, 2010). There are thus considerable methodological variables to consider when attempting to mechanistically link butyrate levels

in faecal matter with circulating concentrations as a conduit of microbiota-influenced behaviours (Topping and Clifton, 2001).

7.3 Butyrate in the brain?

To our knowledge there are no studies on physiological concentrations of butyrate in the brain or CSF. However, given the relatively low levels in peripheral blood, it can be expected that butyrate levels in brain tissue or CSF are extremely low. In fact, a study using dynamic positron emission tomography tracing of radio labelled butyrate in primates found brain uptake to be less than 0.006% and revealed high turnover of butyrate (only 20% remained after 5 minutes) (Kim et al., 2013). In contrast, using ion chromatography two recent studies by Jaming Liu and colleagues found slightly elevated butyrate levels in the brains of mice supplemented with live *Clostridium butyricum*, in a vascular dementia or ischemia mouse model (Liu et al., 2015; Sun et al., 2016). These observations stand to be reproduced as the concentrations of butyrate in wet brain sample reported in these studies ranged from 0.4 to 0.7 µmol/g (i.e. ~mmol/l) and are thus about an order of magnitude higher than concentrations reported in peripheral blood. Notably, it is possible that not all studies have likely detected peak butyrate levels so it is important not to underestimate the potential impact of transient spikes.

7.4 Transporters and Receptors

Evidence for a role of SCFAs in organs outside the digestive system are derived from the fact that specific transmembrane proteins, receptors and transporters, that specifically bind SCFAs and other monocarboxylic acids are expressed by a large variety of cell types, including neurons. Importantly, in order to affect brain function butyrate does not necessarily need to enter the brain but can also indirectly influence processes in the brain by stimulating for example the peripheral nervous system or regulate immune system function. We will thus discuss relevant data on transporters and receptors also in organs and organ systems other than the brain.

7.4.1 Transporters

SCFAs are transported across cell membranes via pH-dependent, H^+-coupled monocarboxylate transporters (MCTs) and sodium-coupled monocarboxylate transporters (SMCTs). More recently, an additional, liver-specific transporter (organic anion transporter 7,

OAT7) was reported to carry out butyrate uptake in hepatocytes (Shin et al., 2007). Both protein families, MCTs and SMCTs, also transport other monocarboxylates, such as pyruvate and lactate, as well as ketone bodies like DHB. Out of the four functional MCTs and two SMCTs, butyrate is a substrate of transporters MCT1 (encoded by the *Slc16a1* gene) and SMCT1 (encoded by the *Slc5a8* gene) (Vijay and Morris, 2014). In the gut, these transporters are located at the apical (i.e. luminal) side of the epithelial cells and their expression is dynamically upregulated specifically in response to luminal butyrate concentrations (Cuff et al., 2002) via the NF-kappaB signalling pathway (Borthakur et al., 2008). In addition, both of these proteins are expressed in multiple tissues and cell types, including kidney, brain and colonic dendritic cells (Ganapathy et al., 2008; Gupta et al., 2006; Kim et al., 2014). Interestingly, in the brain, SMCT1 is found predominantly on neurons, while MCT1 is found mainly on astrocytes (Vijay and Morris, 2014) but also microglia (Moreira et al., 2009) and oligodendrocytes (Lee et al., 2012).

However, more recently MCT4 was suggested to be a high-affinity butyrate transporter, at least in gut epithelial cells (Kekuda et al., 2013). In the brain, MCT4 seems to be exclusively expressed by astrocytes although the implications of this compartmentalisation remain to be defined (Pellerin et al., 2005). Under physiological conditions MCTs and SMCTs are important for shuttling lactate and ketone bodies from astrocytes to neurons for energy metabolism (Martin et al., 2006), but these transporters also have critical roles in the brain for drug delivery and are blocked as off-targets by non-steroidal anti-inflammatory drugs (Vijay and Morris, 2014), and are clearly necessary in mediating the direct effects of butyrate in the brain through uptake into neurons and glia cells from the circulation. In fact, to cross the blood brain barrier (BBB) butyrate-transporting transmembrane proteins must also be expressed by endothelial cells. At least for MCT1 this has been shown repeatedly (Bergersen et al., 2002; Gerhart et al., 1997; Pierre et al., 2000).

However, there is no data available for the SMCTs. Studies using isotope-labelled butyrate were able to demonstrate carrier-mediated uptake of butyrate and other monocarboxylates into the brain as far back as the 1970s (Oldendorf, 1973; Sarna et al., 1979), although more recently this capacity was demonstrated to be limited (Kim et al., 2013). Further evidence for the ability of butyrate to cross the BBB comes from the fact that oral butyrate induces a dose-dependent increase in neuronal and glial nuclear histone H3 acetylation mice (Minamiyama et al., 2004), due to its potential to inhibit histone deacetylation (see Section 8). Interestingly, butyrate itself can regulate BBB integrity: a recent study by Braniste and colleagues at the Karolinska Institute (2014) showed increased permeability of the BBB in germ-free mice, which are lacking detectable levels of butyrate. Monoassociation of these mice with the butyrate-producing bacterium (*Clostridium tyrobutyricum*), as well as oral sodium butyrate

administration (1000mg/kg for 3 days) could reinstate BBB integrity by increasing tight junction protein expression. This could not be achieved by administration *Bacteroides thetaiotaomicron,* producing mainly acetate and propionate, suggesting that indeed fermentation-derived butyrate supported BBB permeability (Braniste et al., 2014). However, from this study it is still unclear if the effects of fermentation-derived butyrate on BBB function are direct or mediated by other cellular systems.

7.4.2 Receptors

In addition to transmembrane transport, butyrate and other SCFAs can influence intracellular signalling in the gut and remote organs by binding to cell surface receptors (Bolognini et al., 2016). So far, four G protein-coupled receptors (GPCRs) have been found to be activated by butyrate: GPR43 (now renamed to free fatty acid receptor 2, **FFAR2**), GPR41 (now **FFAR3**), **GPR109a** and GPR164 (now **OR51E1**, Olfr558 in mice) (**Table 1**). Though with somewhat different affinities, FFAR2 and FFAR3 are activated by all three major SCFAs (Ulven, 2012; Yonezawa et al., 2013), while GPR109a (now renamed to hydroxycarboxylic acid receptor 2, HCAR2 or HCA2) is activated by butyrate, monomethyl fumarate and the B3 vitamin niacin, which gave the encoding gene *niacin receptor 1* (*Niacr1)* its initial name (Singh et al., 2014; Tang et al., 2008). OR51E1 binds butyrate, 3- and 4-methyl valeric acids and nonanoic acid (Priori et al., 2015).

In addition, several synthetic agonists and antagonists have been identified to pharmacologically target FFAR2 and FFAR3 more specifically (Ulven, 2012; Yonezawa et al., 2013). Interestingly, D-β-hydroxybutyrate (DHB) appears to be an endogenous antagonist of FFAR3 (Kimura et al., 2011) but an agonist of HCAR2 (Offermanns and Schwaninger, 2015; Taggart et al., 2005), although evidence on DHB antagonism at FFAR3 in the sympathetic nervous system is inconsistent (López Soto et al., 2014; Won et al., 2013). Of note, there is another GPCR, OLFR78, which is expressed in blood vessels to regulate blood pressure and is activated by acetate and propionate but not by butyrate (Pluznick et al., 2013). Since all these receptors are activated by SCFAs in higher micromolar to millimolar concentrations it is unlikely that they will be activated by butyrate or propionate in organs other than the colon or liver, as peripheral blood usually carries only very low micromolar amounts of these SCFAs.

Table 1: Cellular receptors for butyrate. Form: Formate, Ac: Acetate, Prop: Propionate, But: butyrate, Val: Valerate (Pentanoate, C5), Capr: Caproate (Hexanoate, C6). *Note that there is conflicting evidence for D-β-hydroxybutyrate (DHB) regarding agonist/antagonist status at FFAR3. For further synthetic ligands (agonists and antagonists) for FFAR2 and FFAR3 see (Ulven, 2012), for HCAR2 see http://www.uniprot.org/uniprot/Q8TDS4#function .

Receptor	Gene symbol	Gene name	Other Aliases	Ligands	Potency rank order
FFAR2	*FFAR2*	*free fatty acid receptor 2*	GPR43, GPCR43, FFA2	Form, Ac, Prop, But, Val	Ac ~ Prop > But > Val > Form
FFAR3	*FFAR3*	*free fatty acid receptor 3*	GPR41, GPCR41, FFA3, FFA3R	Ac, Prop, But, Capr, DHB*	Prop ~ But > Val > Ac > Capr
HCAR2	*HCAR2*	*hydroxycarboxylic acid receptor 2*	GPR109a, HCA2, NIACR1, PUMAG, HM74a/b	But, monomethyl fumarate, nicotinic acid (niacin/Vit. B3), DHB	n.d.
OR51E1	*OR51E1* (human) *Olfr558* (mouse)	*olfactory receptor family 51 subfamily E member 1* (human) *olfactory receptor 558* (mouse)	GPR164, POGR, Olfr558	But, 3-/4-methyl valeric acids, nonanoic acid	n.d.

However, the SCFA receptors are implicated in various physiological processes due to their localisation on different cell types. FFAR2 and FFAR3 are located at various cell types, most prominently on enteroendocrine cells, where they, together with OR51E1 (Priori et al., 2015), stimulate secretion of glucagon-like peptide-1 (GLP-1), peptide YY (PYY), and other peptide hormones to regulate appetite and energy homeostasis (Byrne et al., 2015; Nøhr et al., 2013; Sleeth et al., 2010; Yadav et al., 2013) and to potentially improve type 2 diabetes features (Puddu et al., 2014). In contrast, SCFA-induced serotonin release from enterochromaffin cells appears not to be mediated by FFARs (Karaki et al., 2006; Tazoe et al., 2009), but OR51E1 may be involved (Priori et al., 2015).

They are also frequently found on adipocytes, where they regulate adipogenesis and lipolysis (Ang and Ding, 2016; Yonezawa et al., 2013), although there are conflicting reports in the literature as to the exact mechanisms of this effect (Rumberger et al., 2014; Taggart et al., 2005). Notably, at least FFAR3 and HCAR2 expression is regulated by DNA methylation (Remely et al., 2014; Thangaraju et al., 2009) and FFAR3 has been shown to be increased in blood leukocytes in obese and type-II-diabetes patients, potentially leading to reduced satiety signalling via SCFA-induced leptin production in adipocytes (Remely et al., 2014). Moreover, while both receptors are well-established regulators of the immune response FFAR2 appears to be more prevalently present on several different immune cells compared to FFAR3 (Kim et al., 2014). In fact, FFAR2/GPR43 has been found on neutrophils, monocytes, and more recently also T-regulatory cells (T_{regs}) (Smith et al., 2013), arguing for a strong immune-modulatory effect of SCFAs (also see section 9). Moreover, norepinephrinergic sympathetic neurons express both, FFAR2 and FFAR3, and binding of these receptors by SCFAs and DHB enhanced (propionate) or suppressed (DHB) norepinephrine release and sympathetic nervous system activity via an intracellular G-protein (Gβγ) – Phospholipase C (PLCβ) – mitogen-activated kinase (MAPK) pathway or by voltage-dependent inhibition of N-type Ca(2+) channels in sympathetic neurons (Kimura et al., 2011; López Soto et al., 2014; Won et al., 2013). Importantly, Lal et al discovered that butyrate could directly stimulate afferent fibres of the vagus nerve, and while several pathways were ruled out without finding a mechanism or target, receptors were not investigated in this study (Lal et al., 2001). As FFAR3 was found to be expressed in the mouse brainstem vagal ganglion (Nøhr et al., 2015), it is possible that this effect was mediated by butyrate receptors. Together, these data further establish the role of these diet-depended metabolites in systemic signalling.

Notably, HCAR2 is expressed in the mammalian brain, e.g. in rodent hypothalamic neurons and in cattle CNS (Fu et al., 2015; Rezq and Abdel-Rahman, 2016; Titgemeyer et al., 2011), and its signalling has well established effects in brain function. As such, HCAR2 is

upregulated in microglia within the substantia nigra of Parkinson's disease patients (Wakade et al., 2014) and mediates the anti-neuroinflammatory, neuroprotective effects of the recently approved anti-multiple sclerosis (MS) drug dimethyl fumarate by converting microglia from a pro-inflammatory to a neuroprotective phenotype (Chen et al., 2014; Offermanns and Schwaninger, 2015; Parodi et al., 2015). For example, activation of HCAR2 by DHB has also been shown to enhance learning and memory by increasing protein synthesis and gap junctional neuronal communication and may play a role in DHB-mediated neuroprotection during neuronal disorders (Zou et al., 2009). In contrast to the well-studied functions of FFAR2 and FFAR3 in the gut and peripheral organs, it is thought that FFARs are expressed in the brain. However, most evidence supports their expression in the brain only at very low levels (Brown et al., 2003; Nilsson et al., 2003; Nøhr et al., 2013). Whether their expression is inducible by butyrate administration or other experimental paradigms has not been tested.

In conclusion, SCFA receptors are important regulators of immune function, including neuroinflammation, host energy metabolism and endocrine regulation of physiology and behaviour. However, under physiological conditions at non-intestinal locations it is unlikely that butyrate or propionate are directly involved in these processes through activating the respective receptors and other ligands like niacin or DHB might be more important in this regard. Yet it is important to note, differential microbial composition and activity may still influence receptor activation via other pathways such as tryptophan metabolism, which is an essential precursor for niacin synthesis in the host liver. In addition, pulsed peak concentrations of SCFAs, especially acetate might be much higher than experimentally determined so far due to technical constraints. Future research, determining SCFA concentrations in the (human) brain after a fibre-reach meal may confirm or rule out the contribution of SCFAs to directly influence brain function.

8 Butyrate and lysine acetylation

8.1 Butyrate as an HDAC inhibitor

Histone deacetylases (HDACs or KDACs) are a family of proteins catalysing the removal of acetyl groups from lysine ('K') residues within a peptide chain. Acetylation of lysine in proteins is an important mechanism of intracellular signalling (Spange et al., 2009) and is most well-known to be occurring on nucleosomal histone proteins, where acetylation of the histone tails is associated with activation of transcription (**Fig. 2A**). More recently, acetylation of lysines has been initially found in more than 1700 proteins (Choudhary et al., 2009), a number that has since grown to over 4000 in mouse (Baeza et al., 2016). Post-translational acetylation regulates (i.e. activates enzymes, provides bindings sites for other proteins,

and/or induces structural change in proteins and DNA) diverse cellular functions, especially in energy metabolism as 63% of mitochondria-localised proteins contain acetylation sites (Baeza et al., 2016), rivalling the much longer-known regulation of proteins by phosphorylation (Norvell and McMahon, 2010; Zhao et al., 2010).

The HDAC family can further be sub-grouped into five classes (I, IIa, IIb, III, IV) (Fischer et al., 2010). While class I HDACs are mainly localized to the nucleus to deacetylate histone and transcription factors, class IIb HDACs are mainly found in the cytoplasm, where HDAC6 for example deacetylates tubulin. Class IIa HDACs are known to shuttle between cytoplasm and nucleus, depending on their phosphorylation status. As HDACs are such important regulators of multiple cellular functions, they need to be under tight control themselves. The activity of HDAC proteins can be regulated by post-translational modifications, including phosphorylation, sumoylation, ubiquitylation and also acetylation (Brandl et al., 2009; Luo et al., 2009). However, they can also be regulated by endogenous metabolites and synthetic compounds. Prominently, the sirtuins, a group of seven (in mammals) lysine deacetylases that make up class III HDACs and are found in the nucleus, cytoplasm and also in mitochondria, are dependent on the co-factor nicotinamide adenine dinucleotide (NAD^+). To date, a wide array of natural and synthetic inhibitors of HDACs of all 5 classes have been described, with a strong focus on classes I and IIa and IIb as inhibition of these HDACs produced most of the beneficial effects on host metabolism and brain health associated with HDAC inhibition.

Butyrate was found as one of the very first endogenous substances to inhibit HDAC activity, preferentially of classes I, IIa (Cleophas et al., 2016; Davie, 2003). Interestingly, butyrate was first discovered as a somatostatic agent in various cancer cells that promotes differentiation, while studying the effects of fatty acids as antimicrobial food additives on mammalian cell lines (Ginsburg et al., 1973; Leder and Leder, 1975; Prasad and Sinha, 1976). When investigating the molecular mechanism of this effect, butyrate was found to increase histone acetylation (Riggs et al., 1977), which was later attributed to inhibition of HDAC enzymes (Candido et al., 1978; Sealy and Chalkley, 1978). More recently, also the related ketone body DHB was found to have systemic HDAC inhibitory effects, potentially related to beneficial health effects of autophagy, the removal of surplus or damaged intracellular components, associated with intermitted fasting, caloric restriction or a ketogenic diet (Shimazu et al., 2013).

While significantly less potent than artificially designed synthetic compounds, butyrate appears to be the most potent inhibitor among natural compounds investigated so far and especially the most potent SCFA in this regard (**Fig. 2B**) (Gilbert et al., 2006). Interestingly, also a connection between feeding of prebiotic lactulose and caecal tissue histone

acetylation could be established in piglets, suggesting that increased butyrate production in the presence of prebiotics may indirectly alter host histone acetylation at least in gut tissues (Kien et al., 2008).

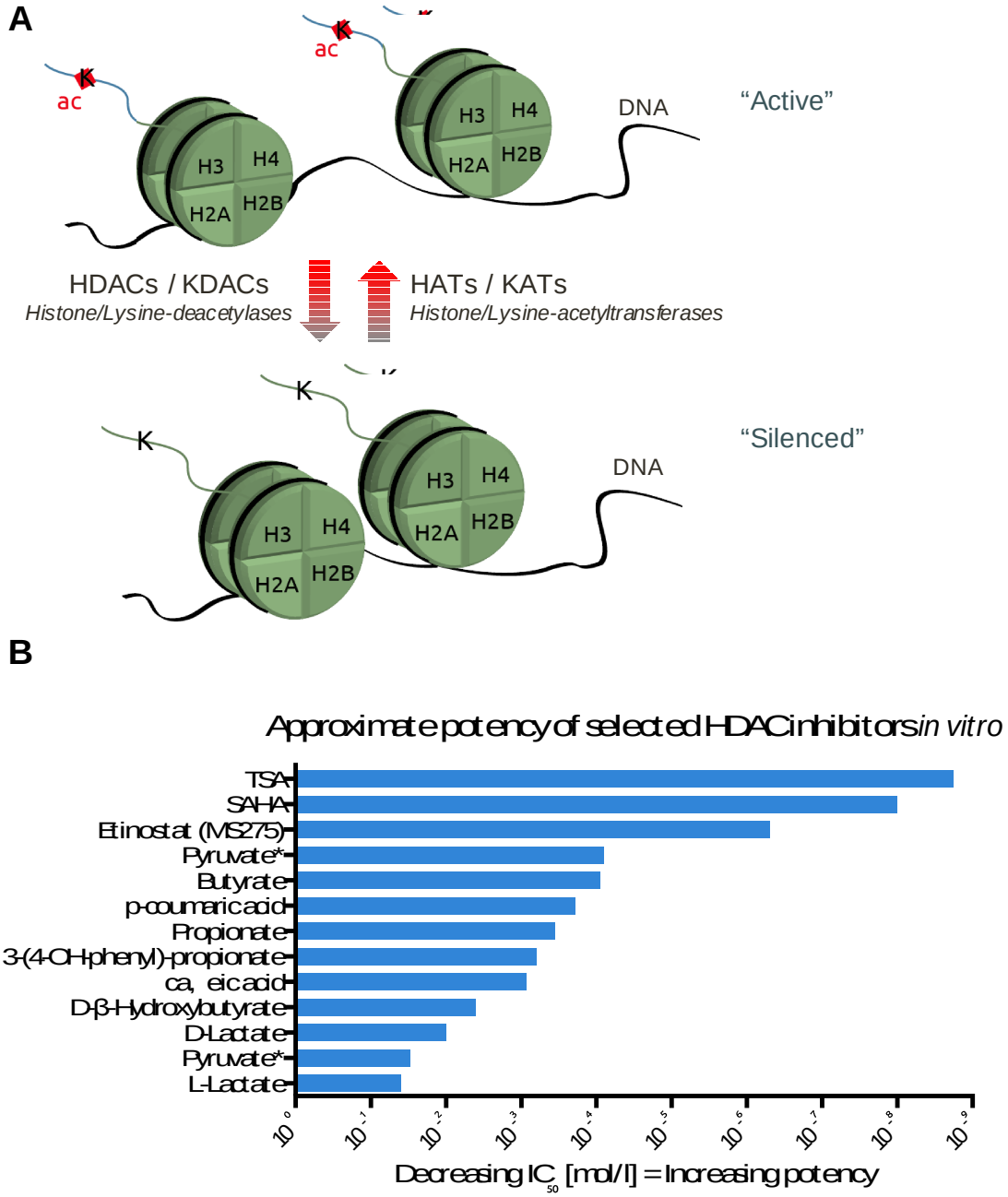


***Fig. 2**: **(A)** Histone acetylation contributes to the formation of open, active chromatin (euchromatin). Acetylation of the lysine (K) tails of the 4 nucleosomal histones (mainly histones H3 and H4) is catalysed by the opposing activity of histone/K-lysine acetyltransferases (HATs/KATs) and histone/K-lysine deacetylases (HDACs/KDACs). **(B)** Approximate IC50 values for different synthetic and natural or endogenous HDAC inhibitors, based on in vitro studies. Note that for pyruvate different values found can be found in the literature. TSA: Trichostatin A, SAHA: suberanilohydroxamic acid*

8.2 Dynamics of neuronal histone acetylation and neurological disorders

8.2.1 Histone acetylation during Long-term memory

Nobel Laurete Eric Kandel's work in *Aplysia*, showed that synthesis of new proteins is necessary for long-term changes in synaptic plasticity and learning (Castellucci et al., 1989; Dale et al., 1987; Montarolo et al., 1986). These findings then led to early studies on the dynamics of histone acetylation during and after a learning task (Levenson et al., 2004) in rodents, which laid the foundation for the emerging field of neuroepigenetics (Sweatt, 2013). This first study by Levenson et al. (2004) also used butyrate for the first time to artificially elevate histone acetylation in the brain during a critical phase of memory formation and found an enhancement of long-term potentiation (LTP) and contextual fear memory (Levenson et al., 2004).

Since then, a multitude of studies have been performed using the HDAC inhibitor butyrate (mostly as sodium salt) to alter brain function in healthy animals or in order to ameliorate detrimental effects in models of cognitive or neurological dysfunction. **Table 2** gives a detailed, but non-exhaustive, overview of several *in vivo* studies using either butyrate, 4-phenylbutyrate, butyrate in conjunction with other SCFAs, or administering butyrate-producing bacteria to modulate brain function, behaviour, or non-neuronal brain components such as the blood brain barrier and glial cells *in vivo*.

In summary, butyrate has been shown to be effective in the brain in at least three major areas. Firstly, based on the findings by Levenson et al. (2004), sodium butyrate was used to facilitate neuronal plasticity, long-term memory formation or LTP (Lattal et al., 2007; Vecsey et al., 2007), e.g. by transforming short-term memory into long-term memory (Haettig et al., 2011; Intlekofer et al., 2013) or by mimicking the beneficial effects of environmental enrichment (Fischer et al., 2007). This memory enhancing feature of high-dosed systemic or locally injected butyrate has also been used for neuroprotection or to restore cognitive function in experimental models of neurodegeneration or cognitive impairment (Ferrante et al., 2003; Fischer et al., 2007; Govindarajan et al., 2011; Kim et al., 2009, 2007; Ryu et al., 2005). To this category also belong other neurodegenerative diseases, including Huntington's disease (Ferrante et al., 2003; Gardian et al., 2005), Parkinson's disease (Sharma et al., 2015), amyotrophic lateral sclerosis (Ryu et al., 2005), and ataxias (Chou et al., 2011), where butyrate exhibits beneficial neuroprotective effects and restoration of brain functions other than memory (**Table 2**). Moreover, butyrate also facilitates neuronal plasticity induced by drugs of abuse, such as cocaine or amphetamine (e.g. Febo et al., 2009; Kalda et al., 2007; Kumar et al., 2005; Sanchis-Segura et al., 2009; Schroeder et al., 2008; Shen et al., 2008) (**Table 2**).

To understand why butyrate has these cognitive enhancement effects during long-term memory formation, it is necessary to consider the molecular mechanisms of this process. Together with the establishment of different phases of memory formation that depend on the strength of synaptic input, the need for *de novo* synthesis of proteins and hence transcriptional regulation specifically during long-term memory formation or late LTP has sparked the generation of the synaptic tagging and capturing hypothesis (Frey and Morris, 1997; Redondo and Morris, 2011; Viosca et al., 2007). In order to facilitate memory and to transform weak sub-threshold stimulation into long-term memory or late LTP, HDAC inhibition must "short-cut" this cellular cascade and activate nuclear gene expression even in the absence of a strong stimulus (**Fig. 3**) (Stilling and Fischer, 2011).

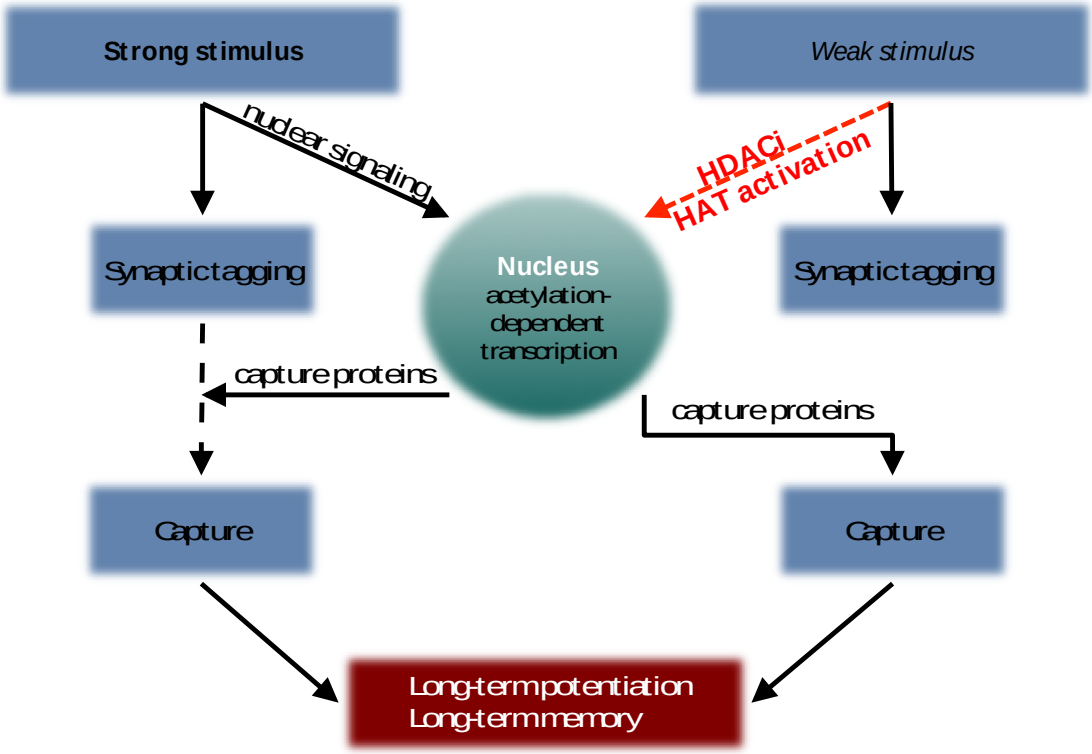


Fig. 3: *Diagram depicting the events occurring in the synaptic tagging and capturing model and how this pathway can be "short-cut" by the use of HDAC inhibitors (HDACi) or HAT activators, both promoting histone acetylation and nuclear gene expression necessary for long-term memory*

It is worth noting that all present evidence that butyrate acts on memory systems via its HDAC inhibitory function is indirect. To prove this beyond doubt would require knockout, knockdown, or pre-inhibition of all HDACs that are inhibited by butyrate, which by itself would either, in the worst-case scenario, be lethal to the cell population (or animal) studied or, in the best-case scenario, result in memory/LTP enhancement when tested. Butyrate should then not add to the enhancement any further – however ceiling effects might confound this approach. Alternatively, systematic elimination of alternative mechanisms of action, such as receptor activation, would be

necessary. While we are not aware of any such systematic studies, we believe that the high reproducibility and wealth of studies showing an effect on histone acetylation, *in vivo* as well as *in vitro,* combined with the philosophy of "Occam's razor", arguing that the most parsimonious explanation is likely the truth, yields sufficient evidence. Hence although the evidence is indirect in most cases, it is convincing to an extend that we can say butyrate facilitates plasticity (at least at the doses mentioned).

8.2.2 Histone acetylation in psychiatric disorders

Psychiatric disorders including depression show important responses to butyrate-induced histone hyperacetylation such as a reduction in depressive like behaviour (Schroeder et al., 2007; Wei et al., 2015) in animal models (**Table 2**). This effect also depends on elevating levels of BDNF in specific brain regions such as the prefrontal cortex (Wei et al., 2015), which is likely due to elevating histone acetylation in the *Bdnf* gene (Intlekofer et al., 2013). In fact, the branched-chain fatty acid valproate, which also has some HDAC inhibitory potential in addition to a variety of other actions, is one of the most widely-used mood stabilisers in clinical practice.

Finally, HDAC inhibition is tightly connected to autism spectrum disorders (ASDs), which are, on a cellular level, characterized by decreased neuro-inhibitory (e.g. GABAergic) signalling. Kratsman et al. (2015) recently showed that butyrate can be used to enhance inhibitory signalling in the BTBR mouse model of autism even at relatively low doses (100mg/kg) that did not induce quantifiable differences in histone acetylation in the prefrontal cortex but still attenuated social deficits associated with this model (Kratsman et al., 2015). In addition, high doses of the monocarboxylic HDAC inhibitors propionate or valproate, administered either systemically to the mother during pregnancy or intraventricular in adulthood were repeatedly shown to induce autistic-like symptoms in animal models (Chomiak et al., 2013; Macfabe, 2012; MacFabe et al., 2011, 2007; Roullet et al., 2013; Thomas et al., 2012), suggesting extreme care has to be given to evaluation of the potential use of SCFAs in treatment of ASDs. Interestingly, however, the prenatal valproate-induced mouse model could also be treated with butyrate or even valproate when given at 4 weeks of age for 5 weeks, which was accompanied by increased histone H3 acetylation in the case of butyrate (Takuma et al., 2014). In the context of the microbiome-gut-brain axis, it is important to note that this model is, along with the neurodevelopmental symptoms, associated with altered composition and activity of the microbiota as well as intestinal inflammation (de Theije et al., 2014a, 2014b).

From **Table 2** it can also be seen investigators used various different doses of butyrate for systemic administration in the range of 100 – 1200 mg/kg. Often these doses were not justified in the studies or were based on previous reports of effectiveness. Only very recently a study by Gagliano et al. (2012) demonstrated negative, stress-like activation of the

hypothalamic-pituitary-brain axis by sodium butyrate in rats at high doses of 1200 mg/kg when injected into the peritoneum, while an equimolar dose of sodium chloride or a butyrate dose of 200 mg/kg did not induce this stress response (Gagliano et al., 2014). Interestingly, this study might also yield an additional mechanism of action for the beneficial effects of high doses of butyrate on memory formation since short-term stress resulting in an acute rise of cortisol levels was shown to facilitate learning (Hurtubise and Howland, 2016). At the same time, the role of HDACs in short-term activation of the HPA axis is unclear. However, compared to physiological conditions, i.e. concentrations of butyrate derived from microbial fermentation in the gut, 200mg/kg and even 100mg/kg (the lowest systemic dose found in **Table 2**) can still be considered very high. Given the low bioavailability of butyrate in the brain (Kim et al., 2013), high doses may have to be used to observe direct effects on HDAC activity in the brain. It seems rather unlikely that doses usually reaching the brain under physiological conditions, even on a high-fibre diet, are high enough to significantly affect histone acetylation. This is an important gap in our understanding of the biology of butyrate that needs to be addressed in well-designed studies. It would also be interesting to determine colonic / faecal levels of butyrate in depressive or other neuropsychiatric patients. Surprisingly, a recent study showed overrepresentation of several bacterial genera that belong to the butyrate producing groups of bacteria in patients with major depressive disorder (Zheng et al., 2016), although SCFA levels were not quantified in this study and the functional causes and consequences of this association still remain to be determined.

Table 1: Selected *in vivo* studies using butyrate, 4-phenylbutyrate or butyrate-producing bacteria to modulate brain physiology and function.

PMID	First Author	Year	Method of Application	Dose [mg/kg]	Duration / Frequency	Experimental Model	Relevance	Main effects
14561870	Ferrante	2003	I.p.	100, 200, 400, 600, and 1200	From 21 days of age throughout lifetime	R6/2 mice	Huntington's disease	Dose-dependent increase in motor performance, lifespan and bodyweight and decreased neuronal atrophy.
14561870	Ferrante	2003	I.p.	1200	2 weeks	R6/2 mice	Huntington's disease	Increased whole brain histone acetylation and increased mitogen-activated protein kinase 1 in the cortex.
15102712	Minamiyama	2004	Drinking water	approx. 600	From 5 weeks of age throughout lifetime	AR-97Q #4–6 mice	Spinal and bulbar muscular atrophy	Increased lifespan and motor performance and increased spinal cord histone acetylation. Amelioration of motor neurons and muscle impairment.
15273246	Levenson	2004	I.p.	1200	Acute	Sprague-Dawley rats		Enhanced contextual fear learning and LTP
16242410	Kumar	2005	I.p.	200	Acute	Sprague-Dawley rats, cotreatment with cocaine	Psychosis and drug abuse	Potentiated locomotor activity and striatal cFos expression and promoter histone acetylation.
15934930	Ryu	2005	I.p.	4-phenylbutyric acid sodium, 400	From 21 days of age throughout lifetime	G93A H1 mice	Amyotrophic lateral sclerosis	Increased lifespan and motor performance. Decreased gross spinal cord atrophy and neuronal loss
15494404	Gardian	2005	I.p.	4-phenylbutyric acid sodium, 100	From 75 days of age throughout lifetime, 6 days/week	N171-82Q mice	Huntington's disease	Increased lifespan, striatal histone acetylation and decreased gross brain atrophy.
16501568	Tsankova	2006	I.p.	200	21 days, 2 times/day	C57Bl/6 mice, after chronic social defeat stress		Decreased depressive-like behavior.
16407196	Ying	2006	I.p.	500 and 1500	From 4 weeks of age, throughout lifetime	Atro-118Q mice	Dentatorubral-pallidoluysian atrophy	Increased lifespan, motor performance and whole brain histone acetylation.
16945350	Schroeder	2007	I.p.	1200	Acute	C57Bl/6J - mice		Increased hippocampal and frontal cortical histone acetylation.
16945350	Schroeder	2007	I.p.	1200	28 days	C57Bl/6J - mice		Decreased depressive-like behaviour.
16945350	Schroeder	2007	I.p.	1200	Acute or 28 days	C57Bl/6J - mice, cotreatment with fluoxetine	Depression	Decreased depressive-like behaviour
17477979	Kalda	2007	I.p.	630	8 days or actute	C57BL/6 mice, cotreatment with amphetamine	Psychosis and drug abuse	Potentiation of amphetamine-induced behavioral sensitization
17477979	Kalda	2007	I.p.	630	6 days, after 8 days of amphetamine administration	C57BL/6 mice, pretreatment with amphetamine	Psychosis and drug abuse	Decreased behavioral sensitization.

21593570	Govindarajan	2011	I.p.	1200	From 14 months of age for 6 weeks	APPPS1-21 mice	Alzheimer's disease	Enhanced contextual fear learning, hippocampal histone acetylation increased expression of genes associated in memory consolidation in the hippocampus.
17477979	Kalda	2007	I.p.	630	Acute	C57BL/6 mice		Increased amphetamine-induced locomotor activity, behavioral sensitization and striatal histone acetylation.
17522015	Bredy	2007	I.p.	1000	Acute	C57BL/6 mice		Increased fear extinction.
17371805	Kim	2007	S.c.	200, 300, 500 and 700	Acutely after pMCAO and 12 hours hereafter	Sprague-Dawley rats with pMCAO	Brain ischemia	Dose-dependent decrease in infarct volume.
17371805	Kim	2007	S.c.	300	Acutely after pMCAO and 12 hours hereafter	Sprague-Dawley rats with pMCAO	Brain ischemia	Increased motor, sensory, and reflex functions and decreased inflammation and apoptosis in the corresponding ischemic brain area.
17907845	Lattal	2007	S.c.	1200	Acute	C57BL/6 mice		Increased fear extinction.
17468743	Fischer	2007	I.p.	1200	4 weeks	C57BL/6 mice		Increased contextual fear learning, spatial learning and hippocampal histone acetylation.
17468743	Fischer	2007	I.p.	1200	4 weeks	CK-p25 Tg mice	Alzheimer's disease	Increased contextual fear learning, spatial learning, hippocampal synaptophysin, neural plasticity factors
17468743	Fischer	2007	I.p.	1200	Acute	C57BL/6 mice		Increased contextual fear learning
17468743	Fischer	2007	I.c.v.	100 ng	Acute	C57BL/6 mice		Increased contextual fear learning
18288092	Schroeder	2007	I.p.	100	10 days	C57BL/6 mice, cotreatment with cocaine	Psychosis and drug abuse	Potentiated cocaine-induced locomotor sensitization.
18288092	Schroeder	2007	I.p.	200	Acute and 10 days	C57BL/6 mice		Increased striatal histone acetylation.
18848971	Shen	2008	I.p.	630	8 days	C57BL/6 mice		Increased striatal ΔFosB expression.
18848971	Shen	2008	I.p.	630	8 days	C57BL/6 mice, cotreatment with amphetamine	Psychosis and drug abuse	Increased amphetamine-induced locomotor sensitization and striatal histone acetylation and CREB phosphorylation.
18799668	Romieu	2008	I.v.	4-phenylbutyrate, 20	7 days	Wistar rats, cotreatment with cocaine	Psychosis and drug abuse	Reduced cocaine self-administration.
18599214	Sun	2008	I.p.	400	Acute	Sprague-Dawley rats, cotreatment with cocaine	Psychosis and drug abuse	Increased cocaine-maintained self-administration.
19549282	Kim	2009	S.c.	300	For 7 or 14 days after pMCAO	Sprague-Dawley rats with pMCAO	Brain ischemia	Increased motor, sensory, and reflex functions and neurogenesis in various brain areas after ischemia

18817816	Zhu	2009	S.c.	1200	For 1 week, 3-4 weeks after the surgery	Ovariectomized female Sprague-Dawley rats, cotreatment with estradiol benzoate	Decreased depressive-like behaviour, Estradiol benzoate was administered due to the ovariectomy
19531374	Dash	2009	I.p.	1200	Acute	C57BL/6 mice	Increased hippocampal histone acetylation.
19531374	Dash	2009	I.p.	1200	For 4 weeks after surgery	C57BL/6 mice with cortical impact injury	Traumatic brain injury	Enhanced learning, memory and contextual fear learning.
19727068	Sanchis-Segura	2009	I.p.	100, 150 and 300	11 days	Swiss-Albino mice, cotreatment with various drugs	Psychosis and drug abuse	Dose-dependent enhancement of cocaine-, ethanol- and morphine-induced locomotor sensitization.
19638299	Febo	2009	I.p.	200	Acute	Long Evans rats, cotreatment with cocaine	Psychosis and drug abuse	Increased cocaine-induced cortico-limbic circuitry activation
19393671	Gundersen	2009	I.p.	1200	3 times in 24 hours	F1 hybrid offspring of 129SvEv and C57Bl/6 mice	Depression and anxiety	Increased depressive-like behaviour and hippocampal histone acetylation.
19393671	Gundersen	2009	I.p.	100	21 days, 2 times/day	F1 hybrid offspring of 129SvEv and C57Bl/6 mice	Depression and anxiety	Decreased hippocampal histone acetylation.
19424149	Guan	2009	I.p.	1200	21 days	C57Bl/6 mice	Enhanced contextual fear learning.
19470462	Stefanko	2009	I.p.	600 and 1200	Acute	Modified Cbp mice	Long-term memory	Increased long-term memory.
20346924	Kwon	2010	I.p.	400 and 1200	Acute	Sprague-Dawley rats	Increased amygdalar histone acetylation.
20407577	Chandrasekar	2010	I.p.	100	3 days	Wistar rats with increased NZF-2b/7ZFMyt1 expression in the nucleus accumbus, cotreatment with cocaine	Psychosis and drug abuse	Decreased cocaine self-administration.
19765687	Malvaez	2010	I.p.	1200	8 days	C57BL/6 mice	Enhanced extinction of cocaine-induced conditioned place preference.
20219993	Gupta	2010	I.p.	N/A	Acute	Sprague-Dawley rats	Enhanced contextual fear learning.
20010553	Kilgore	2010	I.p.	1200	21 and 35 days	APPPS1-21 mice	Alzheimer's disease	Enhanced contextual fear learning.
21597935	Yoo	2011	S.c.	300	2 weeks	C57BL/6 mice	Increased histone acetylation in the dentate gyrus.
20978517	Wang	2011	I.p.	300	Acutely after pMCAO and 12 hours hereafter	Sprague-Dawley rats with pMCAO	Brain ischemia	Attenuation of blood-brain barrier disruption.
21652725	Bonthuis	2011	Drinking water	approx. 1200	20-24 days	Ovariectomized female C57BL/6 mice	Increased whole brain histone acetylation.

21047555	Chou	2011	I.p.	400 and 800	From 2 months of age throughout lifetime	Ataxin-3-Q79 mice	Spinocerebellar ataxia type 3	Increased lifespan, motor performance, histone acetylation in the cerebellum and expression of genes associated with glutamatergic signalling.
21191817	Kim	2011	I.p.	1200	7 days	C57BL/6 mice		Increased hippocampal and cortical histone acetylation and decreased neuronal apoptosis in the subgranular zone of the hippocampus.
21989497	Moretti	2011	I.p.	600	For 7 days, 2 times/day, directly after 7 day d-amphetamine administration	Wistar rats, treated with d-amphetamine for 14 days	Mania and bipolar disorder	Decreased locomotor activity and increased activity of mitochondrial respiratory-chain complexes in the prefrontal cortex, hippocampus, striatum and amygdala.
21224411	Haettig	2011	I.p.	1200	Acute	Mouse (C57Bl/6), CBP functional knockout		HDACi failed to enhance long-term memory in $CBP^{KIX/KIX}$ mice (but enhanced control)
21421011	Reolon	2011	I.p.	1200	Acute	Aged Wistar rats		Increased memory retention.
22457511	Mahan	2012	I.p.	1200	Acute	C57BL/6 mice		Increased contextual fear learning and hippocampal homer1a expression.
22067609	Silva	2012	I.p.	1200	Acute	Wistar rats with neonatal iron overload	Iron overload induced memory impairment	Increased memory retention.
22761874	Yang	2012	I.p.	1200	Acute	Sprague-Dawley rats, cotreatment with sulphasalazine	Psychosis and drug abuse	Abolished the blocked conditioned place aversion memory extinction.
25722691	Xu	2012	I.p.	200	15 days, 4 times/day	Wistar rats, cotreatment with ethanol	Psychosis and drug abuse	Increased ethanol-induced conditioned place preference and htr3a expression.
22452925	Itzhak	2012	I.p.	1200	Acute	nNOS mice		Increased contextual fear learning and amygdalar and hippocampal histone acetylation.
22452925	Itzhak	2012	I.p.	1200	Acute	Parental counterparts of nNOS KO mice (C57BL/6J and 129S1/SvImJ)		Increased extinction of cued fear memory and amygdalar and hippocampal histone acetylation.
22290116	Stafford	2012	I.p.	1200	Acute	C57BL/6 mice		Enhanced fear extinction.
22290116	Stafford	2012	Bilateral dorsal hippocampal injection	55 mM over 1 min at a rate of 0.25 µL per min	Acute	C57BL/6 mice		Increased histone acetylation and cFos levels in the hippocampus and infralimbic cortex.

22290116	Stafford	2012	Unilateral medial infralimbic cortical injection	55 mM over 1 min at a rate of 0.25 µL per min	Acute	C57BL/6 mice		Enhanced fear extinction, did not enhance extinction when injected in prelimic cortex
23615664	Intlekofer	2013	I.p.	N/A	Acute	C57BL/6 mice		Increased subthreshold learning, hippocampal BDNF VI expression and promoter hyperacetylation
23454534	Raybuck	2013	I.p.	1200	Acute	C57BL/6 mice, cotreatment with cocaine	Psychosis and drug abuse	Enhancement of the acquisition and reacquisition of cocaine-induced conditioned place preference and increased histone acetylation in the infralimbic cortex and nucleus accumbens.
23567105	Itzhak	2013	I.p.	1200	Acute	C57BL/6 mice		Increased hippocampal histone acetylation.
23567105	Itzhak	2013	I.p.	1200	Acute	C57BL/6 mice, cotreatment with cocaine	Psychosis and drug abuse	Increased cocaine place preference and surpressed cocaine-induced place preference.
23137698	Harkness	2013	I.p.	630	10 days	B6D2F1/J mice		Potentiated metamphetamine-induced locomoter sensitization.
23137698	Harkness	2013	I.p.	630	Acute	B6D2F1/J mice		Increased histone acetylation in the dorsal and ventral striatum.
23604166	Ploense	2013	I.p.	1000	Acute	Sprague-Dawley rats		Increased cue-induced reinstatement of operant behavior.
23411414	Steckert	2013	I.p.	600	For 7 days, 2 times/day, directly after 7 day d-amphetamine administration	Wistar rats, treated with d-amphetamine for 14 days	Mania and bipolar disorder	Decreased locomotor activity, risk-taking behavior and oxidative stress in various brain regions.
23851624	Valvassori	2013	I.p.	600	For 7 days, 2 times/day, directly after 7 day d-amphetamine administration	Wistar rats, treated with d-amphetamine for 14 days	Mania and bipolar disorder	Increased Krebs cycle enzyme activity.
24212060	Gagliano	2014	I.p.	1200	Acute	Sprague-Dawley rats		Increased stress and anxiety-like behavior and increased hippocampal histone acetylation, 1200mg/kg had negative effects (stress-like response)
24323127	Núñez-Jaramillo	2014	Bilateral insular cortical injection	10 µg	Acute	Wistar rats		Delayed extinction of aversive taste memory, 50 and 100 µg had no effect
25411471	Braniste	2014	Oral gavage	1000	3 days	GF C57BL/6 mice		Blood brain barrier function rescued.

25618518	Wei	2014	I.p.	400	2 times/day	Flinders sensitive line rats	Depression	Decreased depressive-like behaviour, increased prefrontal cortical TET1 and BDNF levels.
24553857	Ji	2014	I.p.	1200	For 3 days during isoflurane exposure	Aged Sprague-Dawley rats with 3 day isoflurane exposure		Increased contextual fear memory, hippocampal histone acetylation, Upregulation of BDNF-TrkB signaling pathway and decreased inflammation and neuronal apoptosis.
25233278	Valvassori	2014	I.p.	500	For 7 days, 2 times/day	Wistar rats		Decreased depressive-like behavior.
25233278	Valvassori	2014	I.p.	500	For 7 days, 2 times/day	Wistar rats with chronic mild stress or maternal deprivation		Decreased activity of tricarboxylic acid cycle enzymes and increased activity of mitochondrial chain complexes.
24753063	Castellano	2014	I.p.	1200	Acute	Long-Evans rats		Increased short-term histone acetylation.
24936215	Kim	2014	S.c.	300	For 7 days after pMCAO	Sprague-Dawley rats with pMCAO	Brain ischemia	Decreased oligodendrocyte loss, inflammation, caspase-3 downregulation and increased oligodendrogenesis.
24607816	Han	2014	I.p.	600	14 days during chronic restraint stress	ICR mice with chronic restraint stress		Decreased depressive-like behaviour and increased hippocampal CREB phosphorylation, histone acetylation in dentate gyrus, CA2/3 and whole hippocampus and increased BDNF in the dentate gyrus and whole hippocampus.
25240644	Takuma	2014	I.p.	1200	5 weeks	ICR mice treated prenatally with valproic acid	Autism spectrum disorder	Increased long-term memory, hippocampal CA1 dendritic spine density and histone acetylation.
24838805	Zhong	2014	I.p.	1200	28 days	C57BL/6 mice		Increased contextual fear learning.
24838805	Zhong	2014	I.p.	1200	28 days prior to isoflurane exposure	C57BL/6 mice exposed to isoflurane		Increased hippocampal CA1 histone acetylation and cFos levels.
24583371	Blank	2014	Bilateral dorsal hippocampal injection	250 mM over 0.5 min at a rate of 2 µL per min	Acute	Wistar rats		Enhanced of inhibitory avoidance memory consolidation.
26013850	Liu	2015	I.p.	840	For 4 weeks, 29 days after 2VO	Sprague-Dawley rats with 2VO	Alzheimer's disease and other types of dementia	Increased spatial memory and hippocampal histone acetylation.
25880762	Castino	2015	I.p.	100	6, 8 and 10 days	Sprague-Dawley rats that underwent 12 days of nicotine self-administration	Psychosis and drug abuse	Increased extinction and attenuation of the reinstatement of nicotine self-administration.
25467060	Varela	2015	I.p.	600	For 7 days, after ICV injection	Wistar rats, with ICV injection of ouabain	Bipolar disorder	Decreased locomotor activity and increased hippocampal and frontal cortical neurotrophic factors.

26064905	Sun	2015	Intragastrical	10	Acutely after BCCAO	ICR mice with BCCAO	Brain ischemia	Decreased neurological deficit and an anti-oxidative and anti-apoptotic effect.
26048426	Sharma	2015	I.p.	150 and 300	For 14 days, 21 days after 6-OHDA treatment	Wistar rats with 6-OHDA administered into the medial forebrain bundle	Parkinson's disease	Dose-dependent increase in motor function, decreased striatal oxidative stress and inflammation and increased striatal BDNF and global histone acetylation.
25433326	Lopes-Borges	2015	I.p.	600	For 7 days, after ICV injection	Wistar rats, with ICV injection of ouabain	Bipolar disorder	Decreased locomotor activity and risk-taking behaviour and increased energetic metabolic alterations in the hippocampus and prefrontal cortex.
25284351	Barichello	2015	Intracisternal	10 mM, 0.5 µL	Acutely after surgery	Wistar rats with intracisternal treatment of Streptococcus pneumoniae	Pneumococcal Meningitis	Decreased memory impairment and increased hippocampal neurotrophic factors.
25041570	Simon-O'Brien	2015	I.p.	600	Acute	Wistar rats, cotreatment with ethanol	Psychosis and drug abuse	Decreased ethanol self-administration.
25041570	Simon-O'Brien	2015	I.p.	600	3 weeks	Long-Evans rats, cotreatment with ethanol	Psychosis and drug abuse	Decreased ethanol intake and preference.
25041570	Simon-O'Brien	2015	I.p.	600	2 days	Wistar rats, cotreatment with ethanol	Psychosis and drug abuse	Increased histone acetylation in various brain areas.
25041570	Simon-O'Brien	2015	I.c.v.	50 or 500 µg	Acute	Wistar rats, cotreatment with ethanol	Psychosis and drug abuse	Decreased ethanol self-administration.
26030851	Erny	2015	Drinking water	approx. 750	4 weeks	GF C57BL/6 mice		Attenuation of defective cortical microglia, given in cocktail with Propionate 25mM, Acetate 67.5mM
24963697	Yoo	2015	S.c.	300	3 weeks	C57BL/6 mice, cotreatment with PEP-1-SIRT2		Increased memory retention and increased cell proliferation and neuroblast differentiation in the dentate gyrus.
26577018	Kratsman	2015	I.p.	100	10 days	BTBR mice	Autism spectrum disorder	Increased sociability, decreased repetitive behaviour and decreased neuronal activation-associated genes and increased HistAc in the prefrontal cortex.
25837444	Blank	2015	I.p.	1200	Acute	Aged Wistar rats		Enhanced of inhibitory avoidance memory consolidation.
26892876	Butchbach	2016	Oral gavage	4-phenylbutyrate, 500	From 4 days of age, throughout lifetime	SMNΔ7 SMA mice	Proximal spinal muscular atrophy	Increased lifespan, number of motor neurons in the lumbar spinal cord and Akt signalling in the spinal cord.

27001149	Jia	2016	I.p.	250	2h prior to sevoflurane exposure and throughout rest experiment	Sprague-Dawley rats, exposed to sevoflurane at P6, P7 and P8		Rescue of deficits in spatial learning, contextual fear learning, hippocampal histone acetylation, BDNF and cFos levels and dendritic spine density
26464112	Blank	2016	Bilateral dorsal hippocampal injection	100 mM over 1 min at a rate of 1 µL per min	Acute	Wistar rats, cotreatment with TrkB antagonist ANA-12		Attenuated decreased inhibitory avoidance memory consolidation induced by ANA-12
27025446	Petry	2016	Bilateral dorsal hippocampal injection	100 mM over 0.5 min at a rate of 2 µL per min	Acute	Wistar rats, cotreatment with hippocampal gastrin-releasing peptide receptor antagonist RC-3095		Attenuated decreased inhibitory avoidance memory consolidation induced by RC-3095 and increased hippocampal BDNF levels.
Studies using butyrate producing bacteria								
25411471	Braniste	2014	Oral gavage	1 × 10^8 CFU Clostridium butyricum	One time 2 weeks prior to experiment	GF C57BL/6 mice		Blood brain barrier function rescued.
26523278	Liu	2015	Oral gavage	1 × 10^6/7/8 CFU Clostridium butyricum	For 6 weeks after rUCCAO	ICR mice with rUCCAO	Vascular dementia	Attenuated spatial learning deficits and decreased hippocampal histopathology and apoptosis, Increased butyrate in the brain and increased faecal microbiota diversity
26733300	Sun	2016	Oral gavage	1 × 10^9 CFU Clostridium butyricum	For 2 weeks prior to BCCAO	ICR mice with BCCAO	Brain ischemia	Decreased neurological deficit and an anti-oxidative and anti-apoptotic effect, Increased butyrate in the brain
27037183	Sun	2016	Oral gavage	1 × 10^8 CFU Clostridium butyricum	For 6 weeks after BCCAO	C57BL/6 mice, 4 week high-fat diet and subsequent streptozotocin administration	Brain ischemia and diabetes	Attenuated spatial learning deficits and hippocampal neuronal injury and apoptosis, Altered gut microbiota composition

9 Butyrate: An effector of immune system, barrier function & tumour growth

Hippocrates famously noted that "*all diseases originate in the gut*". Indeed, the gastrointestinal system offers an integrated interface for regulation of various body functions in health and disease. Strikingly, butyrate has been shown to interact with virtually all of these functions (Canani et al., 2011; Hamer et al., 2008).

As such, the gut epithelium is also the first line of defence against pathogens taken up with the diet. Due to the mutualistic nature of the majority of microbes in the gut, however, the gut epithelium is also the primary interface for host-microbe crosstalk on all levels of interaction (Artis, 2008).

Hence the immune system is trained and regulated by the presence of harmful and beneficial microbes, i.e. their components (e.g. cell wall parts, microbial associated molecular patterns MAMPs as well as microbial and viral DNA and RNA) and products (incl. metabolites). Butyrate has now been established as a potent player mediating this microbe-to-host-immune-system cross talk.

Thus, butyrate and other SCFAs have been shown to strengthen the integrity of the epithelial barrier by upregulating and reorganization of tight junction proteins that connect epithelial cells via various seemingly independent mechanisms including HDAC inhibition as well as activation of the AMP-activated protein kinase (AMPK) and/or lipoxigenase (Bordin et al., 2004; Mariadason et al., 1999, 1997; Ohata et al., 2005; Peng et al., 2009, 2007; Suzuki et al., 2008; Wang et al., 2012). This characteristic is especially important since, unless the vagus nerve is involved, any gut microbe – brain interaction needs to cross at least two barriers, i.e. the gut epithelium and the blood brain barrier, and permeability through both of these barriers has been shown to be affected by the microbiome (Kelly et al., 2015). Moreover, there is accumulating evidence that butyrate has anti-inflammatory potential (Bollrath and Powrie, 2013), and has thus been investigated as a therapeutic agent in inflammatory bowel disorders and colitis (Di Sabatino et al., 2005; Scheppach, 1996; Vieira et al., 2012). Anti-inflammatory activity is achieved through multiple non-mutually exclusive mechanisms: Most prominently, butyrate was found to induce differentiation of T-regulatory cells (Tregs) through a combination of HDAC inhibition and FFAR2 receptor activation (Furusawa et al., 2013; Smith et al., 2013). In addition, it was shown that butyrate appears to deliver a "double-hit" comprising induction of apoptosis of resting and activated $CD4^+$ and $CD8^+$ T cells via inhibition of HDAC1, thereby eliminating one source of inflammation (Zimmerman et al., 2012). Indeed, just recently it could be shown that inflammatory states such as Crohn's disease are associated with a reduction in butyrate-producing bacteria (Takahashi et al., 2016). In contrast, yet also mediated by its HDAC inhibitory role, *in vitro*

butyrate was shown to induce matrix metalloproteinase stromelysin-1 specifically in cytokine-activated intestinal mesenchymal cells, which *increases* inflammatory tissue damage (Pender et al., 2000). This feature provides an important caveat, but needs to be further studied in an *in vivo* context.

An interesting perspective emerges, when we ask the question why the immune system, which evolved to discriminate between self and non-self, harmful and beneficial, responds to butyrate so readily. Why can HDACs be inhibited by butyrate (and to a lesser extent propionate) and why do T_{regs} express receptors for SCFAs? The answer may lie in co-evolution of host immune system and microbial metabolism. The plausible explanation is thus that butyrate, propionate and other microbial metabolites indeed also hold signalling function, providing the immune system with information about the microbiota composition and metabolic activity in the gut. Thereby these immune cells would be able to "sense" the conditions in the gut and may become alert or calmed. For example, a rapid decrease in butyrate concentrations could be indicative of overgrowth of pathogenic bacteria, which needs to be counteracted by the host. As a consequence, also the ability of HDACs to be inhibited by butyrate may have originated through evolutionary pressures in the colon. Since the set of 11 HDACs are the same in the whole host organism, this suggests the possibility of exploiting/targeting this mechanism for pharmacological interventions.

Since depression and other neuropsychiatric illnesses have a pro-inflammatory phenotype (Haapakoski et al., 2016; McKernan et al., 2011) and inflammatory diseases are often associated with depressive symptoms (Miller and Raison, 2016), *vice versa*, butyrate may also be active in these conditions by reducing inflammation. Importantly, the anti-inflammatory feature of butyrate also has fundamental implications for host (brain) ageing (Biagi et al., 2013, 2010; Shimazu et al., 2010), especially in the light of the chronic inflammatory state known as inflamm-ageing (Franceschi et al., 2000) to which many tissues succumb, including the brain (Prenderville et al., 2015). Specifically, butyrate showed anti-inflammatory effects in brain-resident macrophages (microglia), by reducing NF-κB signalling and inducing apoptosis, and thus promoting neuroprotection (Chen et al., 2007; Huuskonen et al., 2004). Furthermore, a recent study by Nakamura et al. (2014) found that long-term feeding of prebiotic fibre ameliorated cognitive decline and had anti-inflammatory, senescence-delaying effects in the SAMP8 mouse model of accelerated ageing (Nakamura et al., 2014). Although this study did not further describe the mechanisms of action for the prebiotics studied, the authors found increased numbers of *Bifidobacteria* and it is likely that SCFA production was increased in parallel (Chen et al., 2008)(also see section 6.1.2), which may, at least in part, be responsible for the observed protective effects. However, due to the very complex metabolic networks within different host tissues a careful assessment of the

advantages and potential disadvantages of SCFAs in ageing and development has to be carried out. For example, a recent study found that GF mice have immature and less active microglia, which could be normalized by adding an SCFA cocktail consisting of acetate, propionate and butyrate to the drinking water (Erny et al., 2015). This suggests that SCFAs not only simply inhibit microglia, but rather support precise tuning to ensure necessary functioning under non-inflammatory conditions. Moreover, a study in *Drosophila* could demonstrate that SCFAs decreased longevity through a connection between metabolism and histone acetylation (Peleg et al., 2016).

10 A role for butyrate in social communication?

Due to their intimate relationship with the host, microbes have been suggested to play important roles in establishing host social behaviours and particularly the evolution and development of mammalian social group living by mutual benefit to the fitness of both host and microbes (Lombardo, 2008; Montiel-Castro et al., 2013, 2014; Stilling et al., 2014a; Troyer, 1984). However, it is not entirely clear how communication between individuals of a certain host species can be influenced by the presence and activity of bacteria.

Butyrate is one of the strongest smells to mammals and humans can detect it at concentrations of about 240 parts per billion (Fazzalari, 1978), possibly via the olfactory receptor OR51E1 (Adipietro et al., 2012). A plausible reason for the good butyrate odour sensitivity of the mammalian nose – usually perceived as a sickening, aversive smell at higher concentrations – is the fact that butyrate is a bacterial product that only occurs under anaerobic conditions, such as biological decomposition, putrefaction or fermentation, which potentially also produce harmful toxins. An alternative, non-mutually exclusive explanation for a high sensitivity to butyrate may be its presence in body odour (Gallagher et al., 2008). It may therefore be used as a social cue to carry information on microbiota composition and activity and thereby indirectly host immune system characteristics, similar to the well-established social signalling function of genetic variability in the major histocompatibility complex (MHC). Peptide ligands of MHC molecules present in urine and sweat are suggested to be social recognition signals that carry information about genetic relatedness and individuality (Boehm and Zufall, 2006) and can be perceived by specialized olfactory receptor neurons in the olfactory epithelium or vomeronasal organ (Overath et al., 2014). Studies in many vertebrates, including humans, have shown that variation at MHC genetic loci influences social behaviour, most prominently mate choice but also cooperative behaviour in social groups (Havlicek and Roberts, 2009; Ruff et al., 2012; Santos et al., 2005; Wedekind et al., 1995). However, other authors have argued that MHC-dependent

olfactory signals are not the only cues that carry information on individuality and genetic variation, and that the “olfactory fingerprint” is more complex (Brown and Schellinck, 1992; Overath et al., 2014). Also variability in volatile carboxylic acid content, possibly including SCFAs, has been shown to reflect genetic MHC-locus variability (Brennan and Kendrick, 2006; Singer et al., 1997), and may in turn be indicative of microbiota composition. In fact, using different strains as well as germ-free rats and colonised germ-free rats, two studies from the early 1990s found that in addition to genetic differences in the MHC cluster, the microbiota is critical for the smell of individuality in urine in rats (Schellinck et al., 1991; Singh et al., 1990). Further evidence for a bacterial influence on social odours and thus social behaviour comes from the study of the microbiota in the scent-producing glands of hyenas. Interestingly, the bacterial communities are more different between individuals from different social groups and more similar within a given group (Theis et al., 2013, 2012). The fact that these microbiota are dominated by fermentative Firmicutes bacteria that produce volatile fatty acids as well as esters, alcohols and aldehydes has prompted the authors to propose the “fermentation hypothesis for chemical communication”, where variation in symbiotic bacteria drives species-, sex- and individual-specific odour variation and thus social communication. The SCFA iso-valeric acid and butyric acid esters were also found in the subauricular scent of the male pronghorn (*Antilocapra americana*) using to mark its territory (Müller-Schwarze et al., 1974). Moreover, the subliminal smell of SCFA valeric acid has been shown to be able to guide social preferences such as decreasing face likeability in human subjects (Li et al., 2007). Thus, it appears plausible that SCFAs like butyrate and other volatile products of microbial fermentation, not only in specialized scent glands but also in other habitats such as the human arm pit, may contribute to chemical communication and convey information about the microbiota composition and hence also genetic information to the interested recipient.

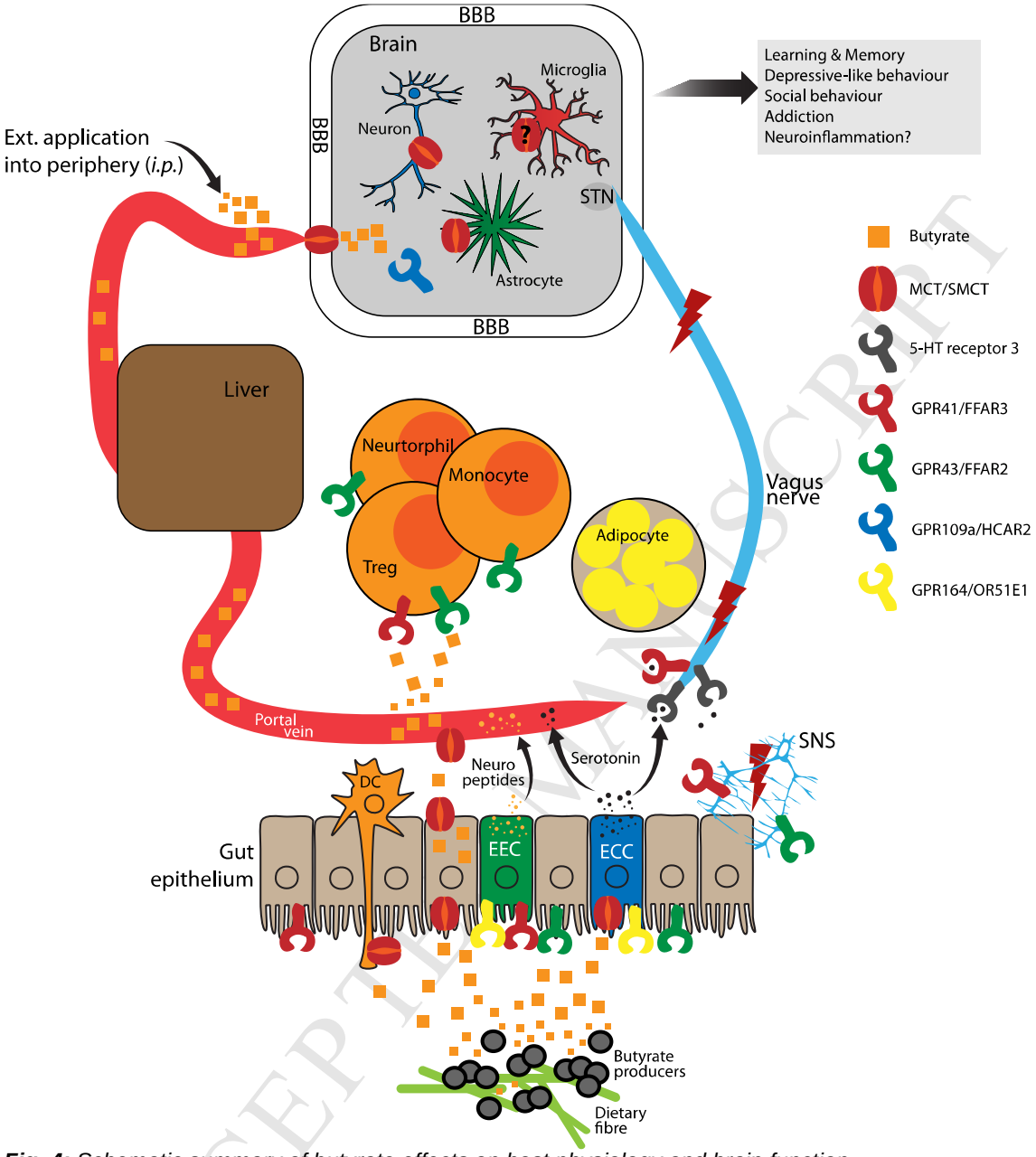


Fig. 4: *Schematic summary of butyrate effects on host physiology and brain function*

11 Conclusions

The current literature points toward mainly positive effects of enhancing production of butyrate and other SCFA in the gut. However, in light of the usually low peripheral concentrations of butyrate and specialised localization of transporters and receptors, it appears very unlikely that butyrate enters the brain in high enough concentrations to exert *direct* molecular effects, such as receptor binding or HDAC inhibition, or to become a feasible energy source under physiological conditions, even when on a high-fibre diet. While we

cannot fully exclude direct effects, the CNS expression of receptors and transporters is thus more likely to be associated with other ligands and transported molecules such as ketone bodies and lactate. In fact there is an unsatisfactory paucity of research on the effects of the constant, low-level exposure to butyrate and other SCFAs, especially during critical neurodevelopmental windows, that needs to be addressed urgently.

Nevertheless, physiological gastrointestinal butyrate can affect the brain. There is ample evidence that production of butyrate by the gut microbiota strongly influences peripheral immune system function, which will in turn shape the brain's immune milieu (Filiano et al., 2015). In addition, butyrate directly affects serotonin and gut hormone release in the enteric nervous system and thereby stimulates the vagus nerve and elicits endocrine signalling, both impacting on brain function. Alternatively, when artificially administered at high concentrations (>100 mg/kg), butyrate acts as a potent drug with well-established, versatile systemic functions. It is thus a valuable neuropharmacological agent, most prominently exploited for its HDAC inhibitory potential.

In summary, butyrate is a functionally extremely versatile molecule (**Fig. 4**), produced by our symbiotic microbes. Host metabolism and immune functions are critically dependent on butyrate as an energy source and potent regulator. This implicates butyrate as a key modifiable mediator of host-microbe crosstalk.

12 Acknowledgements

This publication has emanated from research conducted with the financial support of Science Foundation Ireland (SFI) to the APC Microbiome Institute (Grant Number 12/RC/2273). RMS is supported by the Irish Research Council (IRC) through a Government of Ireland Postdoctoral Fellowship (Grant Number GOIPD/2014/355).

13 References

Adipietro, K.A., Mainland, J.D., Matsunami, H., 2012. Functional Evolution of Mammalian Odorant Receptors. PLoS Genet. 8. doi:10.1371/journal.pgen.1002821

Aitoro, R., Paparo, L., Costanzo, M.D., Nocerino, R., Amoroso, A., Amato, F., Pirozzi, C., Calignano, A., Meli, R., Simeoli, R., Nagler, C., Guandalini, S., Berni, C., 2015. Breast milk butyrate as protective factor against food allergy. Dig. Liver Dis. 47, e274. doi:10.1016/j.dld.2015.07.150

Ang, Z., Ding, J.L., 2016. GPR41 and GPR43 in Obesity and Inflammation – Protective or Causative? Front. Immunol. 7. doi:10.3389/fimmu.2016.00028

Artis, D., 2008. Epithelial-cell recognition of commensal bacteria and maintenance of immune homeostasis in the gut. Nat. Rev. Immunol. 8, 411–420. doi:10.1038/nri2316

Astbury, S., Corfe, B.M., 2012. Uptake and Metabolism of the Short-Chain Fatty Acid Butyrate, a Critical Review of the Literature. Curr. Drug Metab. 13, 815–821. doi:10.2174/138920012800840428

Bach Knudsen, K.E., Serena, A., Canibe, N., Juntunen, K.S., 2003. New insight into butyrate metabolism. Proc. Nutr. Soc. 62, 81–86. doi:10.1079/PNS2002212

Baeza, J., Smallegan, M.J., Denu, J.M., 2016. Mechanisms and Dynamics of Protein Acetylation in Mitochondria. Trends Biochem. Sci. doi:10.1016/j.tibs.2015.12.006

Barcenilla, A., Pryde, S.E., Martin, J.C., Duncan, S.H., Stewart, C.S., Henderson, C., Flint, H.J., 2000. Phylogenetic relationships of butyrate-producing bacteria from the human gut. Appl. Environ. Microbiol. 66, 1654–1661.

Belenguer, A., Duncan, S.H., Calder, A.G., Holtrop, G., Louis, P., Lobley, G.E., Flint, H.J., 2006. Two routes of metabolic cross-feeding between Bifidobacterium adolescentis and butyrate-producing anaerobes from the human gut. Appl. Environ. Microbiol. 72, 3593–3599. doi:10.1128/AEM.72.5.3593-3599.2006

Bergersen, L., Rafiki, A., Ottersen, O.P., 2002. Immunogold cytochemistry identifies specialized membrane domains for monocarboxylate transport in the central nervous system. Neurochem. Res. 27, 89–96.

Biagi, E., Candela, M., Turroni, S., Garagnani, P., Franceschi, C., Brigidi, P., 2013. Ageing and gut microbes: Perspectives for health maintenance and longevity. Pharmacol. Res., SI:Human microbiome and health 69, 11–20. doi:10.1016/j.phrs.2012.10.005

Biagi, E., Nylund, L., Candela, M., Ostan, R., Bucci, L., Pini, E., Nikkïla, J., Monti, D., Satokari, R., Franceschi, C., Brigidi, P., De Vos, W., 2010. Through ageing, and beyond: gut microbiota and inflammatory status in seniors and centenarians. PloS One 5, e10667. doi:10.1371/journal.pone.0010667

Bindelle, J., Buldgen, A., Leterme, P., 2008. Nutritional and environmental consequences of dietary fibre in pig nutrition: a review. Base 12, 69–80.

Boehm, T., Zufall, F., 2006. MHC peptides and the sensory evaluation of genotype. Trends Neurosci. 29, 100–107. doi:10.1016/j.tins.2005.11.006

Bollrath, J., Powrie, F., 2013. Feed Your Tregs More Fiber. Science 341, 463–464. doi:10.1126/science.1242674

Bolognini, D., Tobin, A.B., Milligan, G., Moss, C.E., 2016. The Pharmacology and Function of Receptors for Short-Chain Fatty Acids. Mol. Pharmacol. 89, 388–398. doi:10.1124/mol.115.102301

Bordin, M., D'Atri, F., Guillemot, L., Citi, S., 2004. Histone deacetylase inhibitors up-regulate the expression of tight junction proteins. Mol. Cancer Res. MCR 2, 692–701.

Borthakur, A., Saksena, S., Gill, R.K., Alrefai, W.A., Ramaswamy, K., Dudeja, P.K., 2008. Regulation of monocarboxylate transporter 1 (MCT1) promoter by butyrate in human intestinal epithelial cells: involvement of NF-kappaB pathway. J. Cell. Biochem. 103, 1452–1463. doi:10.1002/jcb.21532

Bourassa, M.W., Alim, I., Bultman, S.J., Ratan, R.R., 2016. Butyrate, Neuroepigenetics and the Gut Microbiome: Can a High Fiber Diet Improve Brain Health? Neurosci. Lett. doi:10.1016/j.neulet.2016.02.009

Brandl, A., Heinzel, T., Krämer, O.H., 2009. Histone deacetylases: salesmen and customers in the post-

translational modification market. Biol. Cell Auspices Eur. Cell Biol. Organ. 101, 193–205. doi:10.1042/BC20080158

Braniste, V., Al-Asmakh, M., Kowal, C., Anuar, F., Abbaspour, A., Tóth, M., Korecka, A., Bakocevic, N., Guan, N.L., Kundu, P., Gulyás, B., Halldin, C., Hultenby, K., Nilsson, H., Hebert, H., Volpe, B.T., Diamond, B., Pettersson, S., 2014. The gut microbiota influences blood-brain barrier permeability in mice. Sci. Transl. Med. 6, 263ra158. doi:10.1126/scitranslmed.3009759

Brennan, P.A., Kendrick, K.M., 2006. Mammalian social odours: attraction and individual recognition. Philos. Trans. R. Soc. B Biol. Sci. 361, 2061–2078. doi:10.1098/rstb.2006.1931

Brown, A.J., Goldsworthy, S.M., Barnes, A.A., Eilert, M.M., Tcheang, L., Daniels, D., Muir, A.I., Wigglesworth, M.J., Kinghorn, I., Fraser, N.J., Pike, N.B., Strum, J.C., Steplewski, K.M., Murdock, P.R., Holder, J.C., Marshall, F.H., Szekeres, P.G., Wilson, S., Ignar, D.M., Foord, S.M., Wise, A., Dowell, S.J., 2003. The Orphan G Protein-coupled Receptors GPR41 and GPR43 Are Activated by Propionate and Other Short Chain Carboxylic Acids. J. Biol. Chem. 278, 11312–11319. doi:10.1074/jbc.M211609200

Brown, R.E., Schellinck, H.M., 1992. Interactions among the MHC, Diet and Bacteria in the Production of Social Odors in Rodents, in: Doty, R.L., Müller-Schwarze, D. (Eds.), Chemical Signals in Vertebrates 6. Springer US, pp. 175–181.

Byrne, C.S., Chambers, E.S., Morrison, D.J., Frost, G., 2015. The role of short chain fatty acids in appetite regulation and energy homeostasis. Int. J. Obes. 39, 1331–1338. doi:10.1038/ijo.2015.84

Canani, R.B., Costanzo, M.D., Leone, L., Pedata, M., Meli, R., Calignano, A., 2011. Potential beneficial effects of butyrate in intestinal and extraintestinal diseases. World J. Gastroenterol. WJG 17, 1519–1528. doi:10.3748/wjg.v17.i12.1519

Candido, E.P., Reeves, R., Davie, J.R., 1978. Sodium butyrate inhibits histone deacetylation in cultured cells. Cell 14, 105–113.

Castellucci, V.F., Blumenfeld, H., Goelet, P., Kandel, E.R., 1989. Inhibitor of protein synthesis blocks long-term behavioral sensitization in the isolated gill-withdrawal reflex of Aplysia. J. Neurobiol. 20, 1–9. doi:10.1002/neu.480200102

Chen, H., Assmann, J.C., Krenz, A., Rahman, M., Grimm, M., Karsten, C.M., Köhl, J., Offermanns, S., Wettschureck, N., Schwaninger, M., 2014. Hydroxycarboxylic acid receptor 2 mediates dimethyl fumarate's protective effect in EAE. J. Clin. Invest. 124, 2188–2192. doi:10.1172/JCI72151

Chen, H.-L., Cheng, H.-C., Wu, W.-T., Liu, Y.-J., Liu, S.-Y., 2008. Supplementation of konjac glucomannan into a low-fiber Chinese diet promoted bowel movement and improved colonic ecology in constipated adults: a placebo-controlled, diet-controlled trial. J. Am. Coll. Nutr. 27, 102–108.

Chen, P.S., Wang, C.-C., Bortner, C.D., Peng, G.-S., Wu, X., Pang, H., Lu, R.-B., Gean, P.-W., Chuang, D.-M., Hong, J.-S., 2007. Valproic Acid and Other HDAC Inhibitors Induce Microglial Apoptosis and Attenuate Lipopolysaccharide- induced Dopaminergic Neurotoxicity. Neuroscience 149, 203–212. doi:10.1016/j.neuroscience.2007.06.053

Chomiak, T., Turner, N., Hu, B., 2013. What We Have Learned about Autism Spectrum Disorder from Valproic Acid. Pathol. Res. Int. 2013, 712758. doi:10.1155/2013/712758

Chou, A.-H., Chen, S.-Y., Yeh, T.-H., Weng, Y.-H., Wang, H.-L., 2011. HDAC inhibitor sodium butyrate reverses transcriptional downregulation and ameliorates ataxic symptoms in a transgenic mouse model of SCA3. Neurobiol. Dis. 41, 481–488. doi:10.1016/j.nbd.2010.10.019

Choudhary, C., Kumar, C., Gnad, F., Nielsen, M.L., Rehman, M., Walther, T.C., Olsen, J.V., Mann, M., 2009. Lysine acetylation targets protein complexes and co-regulates major cellular functions. Science 325, 834–840. doi:10.1126/science.1175371

Clarke, G., Stilling, R.M., Kennedy, P.J., Stanton, C., Cryan, J.F., Dinan, T.G., 2014. Minireview: Gut microbiota: the neglected endocrine organ. Mol. Endocrinol. Baltim. Md 28, 1221–1238. doi:10.1210/me.2014-1108

Cleophas, M.C.P., Crişan, T.O., Lemmers, H., Toenhake-Dijkstra, H., Fossati, G., Jansen, T.L., Dinarello, C.A., Netea, M.G., Joosten, L.A.B., 2016. Suppression of monosodium urate crystal-induced cytokine production by butyrate is mediated by the inhibition of class I histone deacetylases. Ann. Rheum. Dis. 75, 593–600. doi:10.1136/annrheumdis-2014-206258

Collins, M.D., Lawson, P.A., Willems, A., Cordoba, J.J., Fernandez-Garayzabal, J., Garcia, P., Cai, J., Hippe, H., Farrow, J.A., 1994. The phylogeny of the genus Clostridium: proposal of five new genera and eleven new species combinations. Int. J. Syst. Bacteriol. 44, 812–826. doi:10.1099/00207713-44-4-812

Collins, S.M., Surette, M., Bercik, P., 2012. The interplay between the intestinal microbiota and the brain. Nat. Rev. Microbiol. 10, 735–742. doi:10.1038/nrmicro2876

Cryan, J.F., Dinan, T.G., 2012. Mind-altering microorganisms: the impact of the gut microbiota on brain and behaviour. Nat. Rev. Neurosci. 13, 701–712. doi:10.1038/nrn3346

Cuff, M.A., Lambert, D.W., Shirazi-Beechey, S.P., 2002. Substrate-induced regulation of the human colonic monocarboxylate transporter, MCT1. J. Physiol. 539, 361–371.

Cummings, J.H., Macfarlane, G.T., Englyst, H.N., 2001. Prebiotic digestion and fermentation. Am. J. Clin. Nutr. 73, 415s–420s.

Cummings, J.H., Pomare, E.W., Branch, W.J., Naylor, C.P., Macfarlane, G.T., 1987. Short chain fatty acids in human large intestine, portal, hepatic and venous blood. Gut 28, 1221–1227.

Dale, N., Kandel, E.R., Schacher, S., 1987. Serotonin produces long-term changes in the excitability of Aplysia sensory neurons in culture that depend on new protein synthesis. J. Neurosci. Off. J. Soc. Neurosci. 7, 2232–2238.

Davie, J.R., 2003. Inhibition of Histone Deacetylase Activity by Butyrate. J. Nutr. 133, 2485S–2493S.

den Besten, G., van Eunen, K., Groen, A.K., Venema, K., Reijngoud, D.-J., Bakker, B.M., 2013. The role of short-chain fatty acids in the interplay between diet, gut microbiota, and host energy metabolism. J. Lipid Res. 54, 2325–2340. doi:10.1194/jlr.R036012

de Theije, C.G.M., Koelink, P.J., Korte-Bouws, G.A.H., Lopes da Silva, S., Korte, S.M., Olivier, B., Garssen, J., Kraneveld, A.D., 2014a. Intestinal inflammation in a murine model of autism spectrum disorders. Brain. Behav. Immun. 37, 240–247. doi:10.1016/j.bbi.2013.12.004

de Theije, C.G.M., Wopereis, H., Ramadan, M., van Eijndthoven, T., Lambert, J., Knol, J., Garssen, J., Kraneveld, A.D., Oozeer, R., 2014b. Altered gut microbiota and activity in a murine model of autism spectrum disorders. Brain. Behav. Immun. 37, 197–206. doi:10.1016/j.bbi.2013.12.005

De Vuyst, L., Leroy, F., 2011. Cross-feeding between bifidobacteria and butyrate-producing colon bacteria explains bifdobacterial competitiveness, butyrate production, and gas production. Int. J. Food Microbiol., 3rd International Symposium on Propionibacteria and Bifidobacteria: Dairy and Probiotic applications, Oviedo 1-4 June 2010 149, 73–80. doi:10.1016/j.ijfoodmicro.2011.03.003

Di Sabatino, A., Morera, R., Ciccocioppo, R., Cazzola, P., Gotti, S., Tinozzi, F.P., Tinozzi, S., Corazza, G.R., 2005. Oral butyrate for mildly to moderately active Crohn's disease. Aliment. Pharmacol. Ther. 22, 789–794. doi:10.1111/j.1365-2036.2005.02639.x

Duncan, S.H., Holtrop, G., Lobley, G.E., Calder, A.G., Stewart, C.S., Flint, H.J., 2004a. Contribution of acetate to butyrate formation by human faecal bacteria. Br. J. Nutr. 91, 915–923. doi:10.1079/BJN20041150

Duncan, S.H., Louis, P., Flint, H.J., 2004b. Lactate-Utilizing Bacteria, Isolated from Human Feces, That Produce Butyrate as a Major Fermentation Product. Appl. Environ. Microbiol. 70, 5810–5817. doi:10.1128/AEM.70.10.5810-5817.2004

Duncan, S.H., Louis, P., Thomson, J.M., Flint, H.J., 2009. The role of pH in determining the species composition of the human colonic microbiota. Environ. Microbiol. 11, 2112–2122. doi:10.1111/j.1462-2920.2009.01931.x

Egorin, M.J., Yuan, Z.M., Sentz, D.L., Plaisance, K., Eiseman, J.L., 1999. Plasma pharmacokinetics of butyrate after intravenous administration of sodium butyrate or oral administration of tributyrin or sodium butyrate to mice

and rats. Cancer Chemother. Pharmacol. 43, 445–453. doi:10.1007/s002800050922

El Aidy, S., Dinan, T.G., Cryan, J.F., 2014. Immune modulation of the brain-gut-microbe axis. Front. Microbiol. 5, 146. doi:10.3389/fmicb.2014.00146

Erny, D., Hrabě de Angelis, A.L., Jaitin, D., Wieghofer, P., Staszewski, O., David, E., Keren-Shaul, H., Mahlakoiv, T., Jakobshagen, K., Buch, T., Schwierzeck, V., Utermöhlen, O., Chun, E., Garrett, W.S., McCoy, K.D., Diefenbach, A., Staeheli, P., Stecher, B., Amit, I., Prinz, M., 2015. Host microbiota constantly control maturation and function of microglia in the CNS. Nat. Neurosci. 18, 965–977. doi:10.1038/nn.4030

Ewaschuk, J.B., Naylor, J.M., Zello, G.A., 2005. D-lactate in human and ruminant metabolism. J. Nutr. 135, 1619–1625.

Fallingborg, J., 1999. Intraluminal pH of the human gastrointestinal tract. Dan. Med. Bull. 46, 183–196.

Fazzalari, F.A. (Ed.), 1978. Compilation of Odor and Taste Threshold Values Data. ASTM International, 100 Barr Harbor Drive, PO Box C700, West Conshohocken, PA 19428-2959.

Febo, M., Akbarian, S., Schroeder, F.A., Ferris, C.F., 2009. Cocaine-induced metabolic activation in cortico-limbic circuitry is increased after exposure to the histone deacetylase inhibitor, sodium butyrate. Neurosci. Lett. 465, 267–271. doi:10.1016/j.neulet.2009.07.065

Fernandes, J., Su, W., Rahat-Rozenbloom, S., Wolever, T.M.S., Comelli, E.M., 2014. Adiposity, gut microbiota and faecal short chain fatty acids are linked in adult humans. Nutr. Diabetes 4, e121. doi:10.1038/nutd.2014.23

Ferrante, R.J., Kubilus, J.K., Lee, J., Ryu, H., Beesen, A., Zucker, B., Smith, K., Kowall, N.W., Ratan, R.R., Luthi-Carter, R., Hersch, S.M., 2003. Histone deacetylase inhibition by sodium butyrate chemotherapy ameliorates the neurodegenerative phenotype in Huntington's disease mice. J. Neurosci. Off. J. Soc. Neurosci. 23, 9418–9427.

Filiano, A.J., Gadani, S.P., Kipnis, J., 2015. Interactions of innate and adaptive immunity in brain development and function. Brain Res. 1617, 18–27. doi:10.1016/j.brainres.2014.07.050

Fischer, A., Sananbenesi, F., Mungenast, A., Tsai, L.-H., 2010. Targeting the correct HDAC(s) to treat cognitive disorders. Trends Pharmacol. Sci. 31, 605–617. doi:10.1016/j.tips.2010.09.003

Fischer, A., Sananbenesi, F., Wang, X., Dobbin, M., Tsai, L.-H., 2007. Recovery of learning and memory is associated with chromatin remodelling. Nature 447, 178–182. doi:10.1038/nature05772

Flint, H.J., Duncan, S.H., Scott, K.P., Louis, P., 2007. Interactions and competition within the microbial community of the human colon: links between diet and health. Environ. Microbiol. 9, 1101–1111. doi:10.1111/j.1462-2920.2007.01281.x

Forsythe, P., Bienenstock, J., Kunze, W.A., 2014. Vagal Pathways for Microbiome-Brain-Gut Axis Communication, in: Lyte, M., Cryan, J.F. (Eds.), Microbial Endocrinology: The Microbiota-Gut-Brain Axis in Health and Disease, Advances in Experimental Medicine and Biology. Springer New York, pp. 115–133.

Franceschi, C., Bonafè, M., Valensin, S., Olivieri, F., De Luca, M., Ottaviani, E., De Benedictis, G., 2000. Inflamm-aging. An evolutionary perspective on immunosenescence. Ann. N. Y. Acad. Sci. 908, 244–254.

Frey, U., Morris, R.G., 1997. Synaptic tagging and long-term potentiation. Nature 385, 533–536. doi:10.1038/385533a0

Fukumoto, S., Tatewaki, M., Yamada, T., Fujimiya, M., Mantyh, C., Voss, M., Eubanks, S., Harris, M., Pappas, T.N., Takahashi, T., 2003. Short-chain fatty acids stimulate colonic transit via intraluminal 5-HT release in rats. Am. J. Physiol. Regul. Integr. Comp. Physiol. 284, R1269–1276. doi:10.1152/ajpregu.00442.2002

Furusawa, Y., Obata, Y., Fukuda, S., Endo, T.A., Nakato, G., Takahashi, D., Nakanishi, Y., Uetake, C., Kato, K., Kato, T., Takahashi, M., Fukuda, N.N., Murakami, S., Miyauchi, E., Hino, S., Atarashi, K., Onawa, S., Fujimura, Y., Lockett, T., Clarke, J.M., Topping, D.L., Tomita, M., Hori, S., Ohara, O., Morita, T., Koseki, H., Kikuchi, J., Honda, K., Hase, K., Ohno, H., 2013. Commensal microbe-derived butyrate induces the differentiation of colonic regulatory T cells. Nature 504, 446–450. doi:10.1038/nature12721

Fu, S.-P., Liu, B.-R., Wang, J.-F., Xue, W.-J., Liu, H.-M., Zeng, Y.-L., Huang, B.-X., Li, S.-N., Lv, Q.-K., Wang, W.,

Liu, J.-X., 2015. β-Hydroxybutyric acid inhibits growth hormone-releasing hormone synthesis and secretion through the GPR109A/extracellular signal-regulated 1/2 signalling pathway in the hypothalamus. J. Neuroendocrinol. 27, 212–222. doi:10.1111/jne.12256

Gagliano, H., Delgado-Morales, R., Sanz-Garcia, A., Armario, A., 2014. High doses of the histone deacetylase inhibitor sodium butyrate trigger a stress-like response. Neuropharmacology 79, 75–82. doi:10.1016/j.neuropharm.2013.10.031

Gallagher, M., Wysocki, C.J., Leyden, J.J., Spielman, A.I., Sun, X., Preti, G., 2008. Analyses of volatile organic compounds from human skin. Br. J. Dermatol. 159, 780–791. doi:10.1111/j.1365-2133.2008.08748.x

Ganapathy, V., Thangaraju, M., Gopal, E., Martin, P.M., Itagaki, S., Miyauchi, S., Prasad, P.D., 2008. Sodium-coupled Monocarboxylate Transporters in Normal Tissues and in Cancer. AAPS J. 10, 193–199. doi:10.1208/s12248-008-9022-y

Gardian, G., Browne, S.E., Choi, D.-K., Klivenyi, P., Gregorio, J., Kubilus, J.K., Ryu, H., Langley, B., Ratan, R.R., Ferrante, R.J., Beal, M.F., 2005. Neuroprotective effects of phenylbutyrate in the N171-82Q transgenic mouse model of Huntington's disease. J. Biol. Chem. 280, 556–563. doi:10.1074/jbc.M410210200

Gaschott, T., Steinhilber, D., Milovic, V., Stein, J., 2001. Tributyrin, a Stable and Rapidly Absorbed Prodrug of Butyric Acid, Enhances Antiproliferative Effects of Dihydroxycholecalciferol in Human Colon Cancer Cells. J. Nutr. 131, 1839–1843.

Gerhart, D.Z., Enerson, B.E., Zhdankina, O.Y., Leino, R.L., Drewes, L.R., 1997. Expression of monocarboxylate transporter MCT1 by brain endothelium and glia in adult and suckling rats. Am. J. Physiol. 273, E207–213.

Gilbert, K.M., DeLoose, A., Valentine, J.L., Fifer, E.K., 2006. Structure-activity relationship between carboxylic acids and T cell cycle blockade. Life Sci. 78, 2159–2165. doi:10.1016/j.lfs.2005.09.047

Ginsburg, E., Salomon, D., Sreevalsan, T., Freese, E., 1973. Growth inhibition and morphological changes caused by lipophilic acids in mammalian cells. Proc. Natl. Acad. Sci. U. S. A. 70, 2457–2461.

Govindarajan, N., Agis-Balboa, R.C., Walter, J., Sananbenesi, F., Fischer, A., 2011. Sodium butyrate improves memory function in an Alzheimer's disease mouse model when administered at an advanced stage of disease progression. J. Alzheimers Dis. JAD 26, 187–197. doi:10.3233/JAD-2011-110080

Gupta, N., Martin, P.M., Prasad, P.D., Ganapathy, V., 2006. SLC5A8 (SMCT1)-mediated transport of butyrate forms the basis for the tumor suppressive function of the transporter. Life Sci. 78, 2419–2425. doi:10.1016/j.lfs.2005.10.028

Guzmán, M., Blázquez, C., 2004. Ketone body synthesis in the brain: possible neuroprotective effects. Prostaglandins Leukot. Essent. Fatty Acids 70, 287–292. doi:10.1016/j.plefa.2003.05.001

Haapakoski, R., Ebmeier, K.P., Alenius, H., Kivimäki, M., 2016. Innate and adaptive immunity in the development of depression: An update on current knowledge and technological advances. Prog. Neuropsychopharmacol. Biol. Psychiatry 66, 63–72. doi:10.1016/j.pnpbp.2015.11.012

Haettig, J., Stefanko, D.P., Multani, M.L., Figueroa, D.X., McQuown, S.C., Wood, M.A., 2011. HDAC inhibition modulates hippocampus-dependent long-term memory for object location in a CBP-dependent manner. Learn. Mem. 18, 71 –79. doi:10.1101/lm.1986911

Hamer, H.M., Jonkers, D., Venema, K., Vanhoutvin, S., Troost, F.J., Brummer, R.-J., 2008. Review article: the role of butyrate on colonic function. Aliment. Pharmacol. Ther. 27, 104–119. doi:10.1111/j.1365-2036.2007.03562.x

Havlicek, J., Roberts, S.C., 2009. MHC-correlated mate choice in humans: a review. Psychoneuroendocrinology 34, 497–512. doi:10.1016/j.psyneuen.2008.10.007

Holt, R.J., 1971. The esterase and lipase activity of aerobic skin bacteria. Br. J. Dermatol. 85, 18–23.

Hurtubise, J.L., Howland, J.G., 2016. Effects of stress on behavioral flexibility in rodents. Neuroscience. doi:10.1016/j.neuroscience.2016.04.007

Huuskonen, J., Suuronen, T., Nuutinen, T., Kyrylenko, S., Salminen, A., 2004. Regulation of microglial

inflammatory response by sodium butyrate and short-chain fatty acids. Br. J. Pharmacol. 141, 874–880. doi:10.1038/sj.bjp.0705682

Intlekofer, K.A., Berchtold, N.C., Malvaez, M., Carlos, A.J., McQuown, S.C., Cunningham, M.J., Wood, M.A., Cotman, C.W., 2013. Exercise and sodium butyrate transform a subthreshold learning event into long-term memory via a brain-derived neurotrophic factor-dependent mechanism. Neuropsychopharmacol. Off. Publ. Am. Coll. Neuropsychopharmacol. 38, 2027–2034. doi:10.1038/npp.2013.104

Iriki, T., Tamura, K., Ishii, M., Tanaka, H., Miyamoto, T., Onda, K., 2009. Concentrations of ketone body and antidiuretic hormone in cerebrospinal fluid in response to the intra-ruminal administration of butyrate in suckling calves. Anim. Sci. J. 80, 655–661. doi:10.1111/j.1740-0929.2009.00683.x

Jakobsdottir, G., Jädert, C., Holm, L., Nyman, M.E., 2013. Propionic and butyric acids, formed in the caecum of rats fed highly fermentable dietary fibre, are reflected in portal and aortic serum. Br. J. Nutr. 110, 1565–1572. doi:10.1017/S0007114513000809

Kalda, A., Heidmets, L.-T., Shen, H.-Y., Zharkovsky, A., Chen, J.-F., 2007. Histone deacetylase inhibitors modulates the induction and expression of amphetamine-induced behavioral sensitization partially through an associated learning of the environment in mice. Behav. Brain Res. 181, 76–84. doi:10.1016/j.bbr.2007.03.027

Karaki, S., Mitsui, R., Hayashi, H., Kato, I., Sugiya, H., Iwanaga, T., Furness, J.B., Kuwahara, A., 2006. Short-chain fatty acid receptor, GPR43, is expressed by enteroendocrine cells and mucosal mast cells in rat intestine. Cell Tissue Res. 324, 353–360. doi:10.1007/s00441-005-0140-x

Kekuda, R., Manoharan, P., Baseler, W., Sundaram, U., 2013. Monocarboxylate 4 mediated butyrate transport in a rat intestinal epithelial cell line. Dig. Dis. Sci. 58, 660–667. doi:10.1007/s10620-012-2407-x

Kelly, J.R., Kennedy, P.J., Cryan, J.F., Dinan, T.G., Clarke, G., Hyland, N.P., 2015. Breaking down the barriers: the gut microbiome, intestinal permeability and stress-related psychiatric disorders. Front. Cell. Neurosci. 9, 392. doi:10.3389/fncel.2015.00392

Kien, C.L., Chang, J.C., Cooper, J.R., 2000. Butyric Acid Is Synthesized by Piglets. J. Nutr. 130, 234–237.

Kien, C.L., Peltier, C.P., Mandal, S., Davie, J.R., Blauwiekel, R., 2008. Effects of the in vivo supply of butyrate on histone acetylation of cecum in piglets. JPEN J. Parenter. Enteral Nutr. 32, 51–56.

Kim, C.H., Park, J., Kim, M., 2014. Gut Microbiota-Derived Short-Chain Fatty Acids, T Cells, and Inflammation. Immune Netw. 14, 277–288. doi:10.4110/in.2014.14.6.277

Kim, H.J., Leeds, P., Chuang, D.-M., 2009. The HDAC inhibitor, sodium butyrate, stimulates neurogenesis in the ischemic brain. J. Neurochem. 110, 1226–1240. doi:10.1111/j.1471-4159.2009.06212.x

Kim, H.J., Rowe, M., Ren, M., Hong, J.-S., Chen, P.-S., Chuang, D.-M., 2007. Histone deacetylase inhibitors exhibit anti-inflammatory and neuroprotective effects in a rat permanent ischemic model of stroke: multiple mechanisms of action. J. Pharmacol. Exp. Ther. 321, 892–901. doi:10.1124/jpet.107.120188

Kim, S.W., Hooker, J.M., Otto, N., Win, K., Muench, L., Shea, C., Carter, P., King, P., Reid, A.E., Volkow, N.D., Fowler, J.S., 2013. Whole-body pharmacokinetics of HDAC inhibitor drugs, butyric acid, valproic acid and 4-phenylbutyric acid measured with carbon-11 labeled analogs by PET. Nucl. Med. Biol. 40, 912–918. doi:10.1016/j.nucmedbio.2013.06.007

Kimura, I., Inoue, D., Maeda, T., Hara, T., Ichimura, A., Miyauchi, S., Kobayashi, M., Hirasawa, A., Tsujimoto, G., 2011. Short-chain fatty acids and ketones directly regulate sympathetic nervous system via G protein-coupled receptor 41 (GPR41). Proc. Natl. Acad. Sci. 108, 8030–8035. doi:10.1073/pnas.1016088108

Kläring, K., Hanske, L., Bui, N., Charrier, C., Blaut, M., Haller, D., Plugge, C.M., Clavel, T., 2013. Intestinimonas butyriciproducens gen. nov., sp. nov., a butyrate-producing bacterium from the mouse intestine. Int. J. Syst. Evol. Microbiol. 63, 4606–4612. doi:10.1099/ijs.0.051441-0

Knowles, S.E., Jarrett, I.G., Filsell, O.H., Ballard, F.J., 1974. Production and utilization of acetate in mammals. Biochem. J. 142, 401–411. doi:10.1042/bj1420401

Kratsman, N., Getselter, D., Elliott, E., 2015. Sodium butyrate attenuates social behavior deficits and modifies the

transcription of inhibitory/excitatory genes in the frontal cortex of an autism model. Neuropharmacology 102, 136–145. doi:10.1016/j.neuropharm.2015.11.003

Kumar, A., Choi, K.-H., Renthal, W., Tsankova, N.M., Theobald, D.E.H., Truong, H.-T., Russo, S.J., Laplant, Q., Sasaki, T.S., Whistler, K.N., Neve, R.L., Self, D.W., Nestler, E.J., 2005. Chromatin remodeling is a key mechanism underlying cocaine-induced plasticity in striatum. Neuron 48, 303–314. doi:10.1016/j.neuron.2005.09.023

Lal, S., Kirkup, A.J., Brunsden, A.M., Thompson, D.G., Grundy, D., 2001. Vagal afferent responses to fatty acids of different chain length in the rat. Am. J. Physiol. Gastrointest. Liver Physiol. 281, G907–915.

Lattal, K.M., Barrett, R.M., Wood, M.A., 2007. Systemic or intrahippocampal delivery of histone deacetylase inhibitors facilitates fear extinction. Behav. Neurosci. 121, 1125–1131. doi:10.1037/0735-7044.121.5.1125

Leder, A., Leder, P., 1975. Butyric acid, a potent inducer of erythroid differentiation in cultured erythroleukemic cells. Cell 5, 319–322. doi:10.1016/0092-8674(75)90107-5

Lee, Y., Morrison, B.M., Li, Y., Lengacher, S., Farah, M.H., Hoffman, P.N., Liu, Y., Tsingalia, A., Jin, L., Zhang, P.-W., Pellerin, L., Magistretti, P.J., Rothstein, J.D., 2012. Oligodendroglia metabolically support axons and contribute to neurodegeneration. Nature 487, 443–448. doi:10.1038/nature11314

Levenson, J.M., O'Riordan, K.J., Brown, K.D., Trinh, M.A., Molfese, D.L., Sweatt, J.D., 2004. Regulation of Histone Acetylation during Memory Formation in the Hippocampus. J. Biol. Chem. 279, 40545–40559. doi:10.1074/jbc.M402229200

Levine, U.Y., Looft, T., Allen, H.K., Stanton, T.B., 2013. Butyrate-Producing Bacteria, Including Mucin Degraders, from the Swine Intestinal Tract. Appl. Environ. Microbiol. 79, 3879–3881. doi:10.1128/AEM.00589-13

Li, C.-J. (Ed.), 2014. Butyrate: food sources, functions and health benefits. Nova Science Publishers, Inc, New York.

Li, R.W., Li, C.-J., 2014. Enhancing Butyrate Biosynthesis in the Gut for Health Benefits., in: Li, C.-J. (Ed.), Butyrate: Food Sources, Functions and Health Benefits. Nova Science Publishers, Inc, New York.

Liu, J., Sun, J., Wang, F., Yu, X., Ling, Z., Li, H., Zhang, H., Jin, J., Chen, W., Pang, M., Yu, J., He, Y., Xu, J., 2015. Neuroprotective Effects of Clostridium butyricum against Vascular Dementia in Mice via Metabolic Butyrate. BioMed Res. Int. 2015, e412946. doi:10.1155/2015/412946

Li, W., Moallem, I., Paller, K.A., Gottfried, J.A., 2007. Subliminal smells can guide social preferences. Psychol. Sci. 18, 1044–1049. doi:10.1111/j.1467-9280.2007.02023.x

Lombardo, M.P., 2008. Access to mutualistic endosymbiotic microbes: an underappreciated benefit of group living. Behav. Ecol. Sociobiol. 62, 479–497. doi:10.1007/s00265-007-0428-9

López Soto, E.J., Gambino, L.O., Mustafá, E.R., 2014. Free fatty acid receptor 3 is a key target of short chain fatty acid. Channels 8, 169–171. doi:10.4161/chan.28956

Louis, P., Duncan, S.H., McCrae, S.I., Millar, J., Jackson, M.S., Flint, H.J., 2004. Restricted distribution of the butyrate kinase pathway among butyrate-producing bacteria from the human colon. J. Bacteriol. 186, 2099–2106.

Louis, P., Flint, H.J., 2009. Diversity, metabolism and microbial ecology of butyrate-producing bacteria from the human large intestine. FEMS Microbiol. Lett. 294, 1–8. doi:10.1111/j.1574-6968.2009.01514.x

Luo, Y., Jian, W., Stavreva, D., Fu, X., Hager, G., Bungert, J., Huang, S., Qiu, Y., 2009. Trans-regulation of Histone Deacetylase Activities through Acetylation. J. Biol. Chem. 284, 34901–34910. doi:10.1074/jbc.M109.038356

Lyte, M., 2013. Microbial Endocrinology in the Microbiome-Gut-Brain Axis: How Bacterial Production and Utilization of Neurochemicals Influence Behavior. PLoS Pathog 9, e1003726. doi:10.1371/journal.ppat.1003726

Macfabe, D.F., 2012. Short-chain fatty acid fermentation products of the gut microbiome: implications in autism spectrum disorders. Microb. Ecol. Health Dis. 23. doi:10.3402/mehd.v23i0.19260

MacFabe, D.F., Cain, D.P., Rodriguez-Capote, K., Franklin, A.E., Hoffman, J.E., Boon, F., Taylor, A.R., Kavaliers,

M., Ossenkopp, K.-P., 2007. Neurobiological effects of intraventricular propionic acid in rats: Possible role of short chain fatty acids on the pathogenesis and characteristics of autism spectrum disorders. Behav. Brain Res. 176, 149–169. doi:10.1016/j.bbr.2006.07.025

MacFabe, D.F., Cain, N.E., Boon, F., Ossenkopp, K.-P., Cain, D.P., 2011. Effects of the enteric bacterial metabolic product propionic acid on object-directed behavior, social behavior, cognition, and neuroinflammation in adolescent rats: Relevance to autism spectrum disorder. Behav. Brain Res. 217, 47–54. doi:10.1016/j.bbr.2010.10.005

Macfarlane, S., Macfarlane, G.T., 2003. Regulation of short-chain fatty acid production. Proc. Nutr. Soc. 62, 67–72. doi:10.1079/PNS2002207

Mariadason, J.M., Barkla, D.H., Gibson, P.R., 1997. Effect of short-chain fatty acids on paracellular permeability in Caco-2 intestinal epithelium model. Am. J. Physiol. 272, G705–712.

Mariadason, J.M., Catto-Smith, A., Gibson, P.R., 1999. Modulation of distal colonic epithelial barrier function by dietary fibre in normal rats. Gut 44, 394–399.

Martin, P.M., Gopal, E., Ananth, S., Zhuang, L., Itagaki, S., Prasad, B.M., Smith, S.B., Prasad, P.D., Ganapathy, V., 2006. Identity of SMCT1 (SLC5A8) as a neuron-specific Na+-coupled transporter for active uptake of L-lactate and ketone bodies in the brain. J. Neurochem. 98, 279–288. doi:10.1111/j.1471-4159.2006.03878.x

Mayer, E.A., Knight, R., Mazmanian, S.K., Cryan, J.F., Tillisch, K., 2014. Gut microbes and the brain: paradigm shift in neuroscience. J. Neurosci. Off. J. Soc. Neurosci. 34, 15490–15496. doi:10.1523/JNEUROSCI.3299-14.2014

McKernan, D.P., Dennison, U., Gaszner, G., Cryan, J.F., Dinan, T.G., 2011. Enhanced peripheral toll-like receptor responses in psychosis: further evidence of a pro-inflammatory phenotype. Transl. Psychiatry 1, e36. doi:10.1038/tp.2011.37

McOrist, A.L., Miller, R.B., Bird, A.R., Keogh, J.B., Noakes, M., Topping, D.L., Conlon, M.A., 2011. Fecal butyrate levels vary widely among individuals but are usually increased by a diet high in resistant starch. J. Nutr. 141, 883–889. doi:10.3945/jn.110.128504

Miller, A.H., Raison, C.L., 2016. The role of inflammation in depression: from evolutionary imperative to modern treatment target. Nat. Rev. Immunol. 16, 22–34. doi:10.1038/nri.2015.5

Minamiyama, M., Katsuno, M., Adachi, H., Waza, M., Sang, C., Kobayashi, Y., Tanaka, F., Doyu, M., Inukai, A., Sobue, G., 2004. Sodium butyrate ameliorates phenotypic expression in a transgenic mouse model of spinal and bulbar muscular atrophy. Hum. Mol. Genet. 13, 1183–1192. doi:10.1093/hmg/ddh131

Miyoshi, M., Sakaki, H., Usami, M., Iizuka, N., Shuno, K., Aoyama, M., Usami, Y., 2011. Oral administration of tributyrin increases concentration of butyrate in the portal vein and prevents lipopolysaccharide-induced liver injury in rats. Clin. Nutr. 30, 252–258. doi:10.1016/j.clnu.2010.09.012

Montarolo, P.G., Goelet, P., Castellucci, V.F., Morgan, J., Kandel, E.R., Schacher, S., 1986. A critical period for macromolecular synthesis in long-term heterosynaptic facilitation in Aplysia. Science 234, 1249–1254.

Montiel-Castro, A.J., González-Cervantes, R.M., Bravo-Ruiseco, G., Pacheco-López, G., 2013. The microbiota–gut–brain axis: neurobehavioral correlates, health and sociality. Front. Integr. Neurosci. 7, 70. doi:10.3389/fnint.2013.00070

Montiel-Castro, null, Augusto, J., Baez-Yanez, null, Mario, G., Pacheco-Lopez, G., 2014. Social neuroeconomics: the influence of microbiota in partner-choice and sociality. Curr. Pharm. Des. 20, 4774–4783.

Moreira, T.J.T.P., Pierre, K., Maekawa, F., Repond, C., Cebere, A., Liljequist, S., Pellerin, L., 2009. Enhanced cerebral expression of MCT1 and MCT2 in a rat ischemia model occurs in activated microglial cells. J. Cereb. Blood Flow Metab. Off. J. Int. Soc. Cereb. Blood Flow Metab. 29, 1273–1283. doi:10.1038/jcbfm.2009.50

Müller-Schwarze, D., Müller-Schwarze, C., Singer, A.G., Silverstein, R.M., 1974. Mammalian pheromone: identification of active component in the subauricular scent of the male pronghorn. Science 183, 860–862. doi:10.1126/science.183.4127.860

Nakamura, S., Kondo, N., Yamaguchi, Y., Hashiguchi, M., Tanabe, K., Ushiroda, C., Kawahashi-Tokuhisa, M., Yui, K., Miyakoda, M., Oku, T., 2014. Daily Feeding of Fructooligosaccharide or Glucomannan Delays Onset of Senescence in SAMP8 Mice. Gastroenterol. Res. Pract. 2014, 303184. doi:10.1155/2014/303184

Nandedkar, A.K., Kumar, S., 1969. Biosynthesis of fatty acids in mammary tissue. II. Synthesis of butyrate in lactating rabbit mammary supernatant fraction by the reversal of beta-oxidation. Arch. Biochem. Biophys. 134, 563–571.

Nandedkar, A.K., Schirmer, E.W., Pynadath, T.I., Kumar, S., 1969. Biosynthesis of fatty acids in mammary tissue. I. Purification and properties of fatty acid synthetase from lactating-goat mammary tissue. Arch. Biochem. Biophys. 134, 554–562.

Nilsson, N.E., Kotarsky, K., Owman, C., Olde, B., 2003. Identification of a free fatty acid receptor, FFA2R, expressed on leukocytes and activated by short-chain fatty acids. Biochem. Biophys. Res. Commun. 303, 1047–1052.

Nøhr, M.K., Egerod, K.L., Christiansen, S.H., Gille, A., Offermanns, S., Schwartz, T.W., Møller, M., 2015. Expression of the short chain fatty acid receptor GPR41/FFAR3 in autonomic and somatic sensory ganglia. Neuroscience 290, 126–137. doi:10.1016/j.neuroscience.2015.01.040

Nøhr, M.K., Pedersen, M.H., Gille, A., Egerod, K.L., Engelstoft, M.S., Husted, A.S., Sichlau, R.M., Grunddal, K.V., Poulsen, S.S., Han, S., Jones, R.M., Offermanns, S., Schwartz, T.W., 2013. GPR41/FFAR3 and GPR43/FFAR2 as cosensors for short-chain fatty acids in enteroendocrine cells vs FFAR3 in enteric neurons and FFAR2 in enteric leukocytes. Endocrinology 154, 3552–3564. doi:10.1210/en.2013-1142

Norvell, A., McMahon, S.B., 2010. Rise of the rival. Science 327, 964–965. doi:10.1126/science.1187159

Offermanns, S., Schwaninger, M., 2015. Nutritional or pharmacological activation of HCA(2) ameliorates neuroinflammation. Trends Mol. Med. 21, 245–255. doi:10.1016/j.molmed.2015.02.002

Ohata, A., Usami, M., Miyoshi, M., 2005. Short-chain fatty acids alter tight junction permeability in intestinal monolayer cells via lipoxygenase activation. Nutr. Burbank Los Angel. Cty. Calif 21, 838–847. doi:10.1016/j.nut.2004.12.004

Oldendorf, W.H., 1973. Carrier-mediated blood-brain barrier transport of short-chain monocarboxylic organic acids. Am. J. Physiol. 224, 1450–1453.

Overath, P., Sturm, T., Rammensee, H.-G., 2014. Of volatiles and peptides: in search for MHC-dependent olfactory signals in social communication. Cell. Mol. Life Sci. 71, 2429–2442. doi:10.1007/s00018-014-1559-6

Parodi, B., Rossi, S., Morando, S., Cordano, C., Bragoni, A., Motta, C., Usai, C., Wipke, B.T., Scannevin, R.H., Mancardi, G.L., Centonze, D., Kerlero de Rosbo, N., Uccelli, A., 2015. Fumarates modulate microglia activation through a novel HCAR2 signaling pathway and rescue synaptic dysregulation in inflamed CNS. Acta Neuropathol. (Berl.) 130, 279–295. doi:10.1007/s00401-015-1422-3

Payne, A.N., Chassard, C., Zimmermann, M., Müller, P., Stinca, S., Lacroix, C., 2011. The metabolic activity of gut microbiota in obese children is increased compared with normal-weight children and exhibits more exhaustive substrate utilization. Nutr. Diabetes 1, e12. doi:10.1038/nutd.2011.8

Peleg, S., Feller, C., Forne, I., Schiller, E., Sévin, D.C., Schauer, T., Regnard, C., Straub, T., Prestel, M., Klima, C., Schmitt Nogueira, M., Becker, L., Klopstock, T., Sauer, U., Becker, P.B., Imhof, A., Ladurner, A.G., 2016. Life span extension by targeting a link between metabolism and histone acetylation in Drosophila. EMBO Rep. 17, 455–469. doi:10.15252/embr.201541132

Pellerin, L., Bergersen, L.H., Halestrap, A.P., Pierre, K., 2005. Cellular and subcellular distribution of monocarboxylate transporters in cultured brain cells and in the adult brain. J. Neurosci. Res. 79, 55–64. doi:10.1002/jnr.20307

Pender, S.L., Quinn, J.J., Sanderson, I.R., MacDonald, T.T., 2000. Butyrate upregulates stromelysin-1 production by intestinal mesenchymal cells. Am. J. Physiol. Gastrointest. Liver Physiol. 279, G918–924.

Peng, L., He, Z., Chen, W., Holzman, I.R., Lin, J., 2007. Effects of butyrate on intestinal barrier function in a Caco-

2 cell monolayer model of intestinal barrier. Pediatr. Res. 61, 37–41. doi:10.1203/01.pdr.0000250014.92242.f3

Peng, L., Li, Z.-R., Green, R.S., Holzman, I.R., Lin, J., 2009. Butyrate enhances the intestinal barrier by facilitating tight junction assembly via activation of AMP-activated protein kinase in Caco-2 cell monolayers. J. Nutr. 139, 1619–1625. doi:10.3945/jn.109.104638

Peters, S.G., Pomare, E.W., Fisher, C.A., 1992. Portal and peripheral blood short chain fatty acid concentrations after caecal lactulose instillation at surgery. Gut 33, 1249–1252.

Pierre, K., Pellerin, L., Debernardi, R., Riederer, B.M., Magistretti, P.J., 2000. Cell-specific localization of monocarboxylate transporters, MCT1 and MCT2, in the adult mouse brain revealed by double immunohistochemical labeling and confocal microscopy. Neuroscience 100, 617–627.

Pluznick, J.L., Protzko, R.J., Gevorgyan, H., Peterlin, Z., Sipos, A., Han, J., Brunet, I., Wan, L.-X., Rey, F., Wang, T., Firestein, S.J., Yanagisawa, M., Gordon, J.I., Eichmann, A., Peti-Peterdi, J., Caplan, M.J., 2013. Olfactory receptor responding to gut microbiota-derived signals plays a role in renin secretion and blood pressure regulation. Proc. Natl. Acad. Sci. U. S. A. 110, 4410–4415. doi:10.1073/pnas.1215927110

Prasad, K.N., Sinha, P.K., 1976. Effect of sodium butyrate on mammalian cells in culture: a review. In Vitro 12, 125–132.

Prenderville, J.A., Kennedy, P.J., Dinan, T.G., Cryan, J.F., 2015. Adding fuel to the fire: the impact of stress on the ageing brain. Trends Neurosci. 38, 13–25.

Priori, D., Colombo, M., Clavenzani, P., Jansman, A.J.M., Lallès, J.-P., Trevisi, P., Bosi, P., 2015. The Olfactory Receptor OR51E1 Is Present along the Gastrointestinal Tract of Pigs, Co-Localizes with Enteroendocrine Cells and Is Modulated by Intestinal Microbiota. PloS One 10, e0129501. doi:10.1371/journal.pone.0129501

Pryde, S.E., Duncan, S.H., Hold, G.L., Stewart, C.S., Flint, H.J., 2002. The microbiology of butyrate formation in the human colon. FEMS Microbiol. Lett. 217, 133–139. doi:10.1111/j.1574-6968.2002.tb11467.x

Puddu, A., Sanguineti, R., Montecucco, F., Viviani, G.L., 2014. Evidence for the gut microbiota short-chain fatty acids as key pathophysiological molecules improving diabetes. Mediators Inflamm. 2014, 162021. doi:10.1155/2014/162021

Rechkemmer, G., Rönnau, K., von Engelhardt, W., 1988. Fermentation of polysaccharides and absorption of short chain fatty acids in the mammalian hindgut. Comp. Biochem. Physiol. A 90, 563–568.

Redondo, R.L., Morris, R.G.M., 2011. Making memories last: the synaptic tagging and capture hypothesis. Nat Rev Neurosci 12, 17–30. doi:10.1038/nrn2963

Reigstad, C.S., Salmonson, C.E., Rainey, J.F., Szurszewski, J.H., Linden, D.R., Sonnenburg, J.L., Farrugia, G., Kashyap, P.C., 2015. Gut microbes promote colonic serotonin production through an effect of short-chain fatty acids on enterochromaffin cells. FASEB J. Off. Publ. Fed. Am. Soc. Exp. Biol. 29, 1395–1403. doi:10.1096/fj.14-259598

Reineccius, G., Heath, H.B., 2006. Flavor chemistry and technology, 2nd ed. ed. Taylor & Francis, Boca Raton.

Remely, M., Aumueller, E., Merold, C., Dworzak, S., Hippe, B., Zanner, J., Pointner, A., Brath, H., Haslberger, A.G., 2014. Effects of short chain fatty acid producing bacteria on epigenetic regulation of FFAR3 in type 2 diabetes and obesity. Gene 537, 85–92. doi:10.1016/j.gene.2013.11.081

Rezq, S., Abdel-Rahman, A.A., 2016. Central GPR109A Activation Mediates Glutamate-Dependent Pressor Response in Conscious Rats. J. Pharmacol. Exp. Ther. 356, 457–466. doi:10.1124/jpet.115.229146

Rhee, S.H., Pothoulakis, C., Mayer, E.A., 2009. Principles and clinical implications of the brain-gut-enteric microbiota axis. Nat. Rev. Gastroenterol. Hepatol. 6, 306–314. doi:10.1038/nrgastro.2009.35

Riggs, M.G., Whittaker, R.G., Neumann, J.R., Ingram, V.M., 1977. n-Butyrate causes histone modification in HeLa and Friend erythroleukaemia cells. Nature 268, 462–464.

Rios-Covian, D., Gueimonde, M., Duncan, S.H., Flint, H.J., de los Reyes-Gavilan, C.G., 2015. Enhanced butyrate formation by cross-feeding between Faecalibacterium prausnitzii and Bifidobacterium adolescentis. FEMS Microbiol. Lett. 362. doi:10.1093/femsle/fnv176

Rivière, A., Gagnon, M., Weckx, S., Roy, D., De Vuyst, L., 2015. Mutual Cross-Feeding Interactions between Bifidobacterium longum subsp. longum NCC2705 and Eubacterium rectale ATCC 33656 Explain the Bifidogenic and Butyrogenic Effects of Arabinoxylan Oligosaccharides. Appl. Environ. Microbiol. 81, 7767–7781. doi:10.1128/AEM.02089-15

Roullet, F.I., Lai, J.K.Y., Foster, J.A., 2013. In utero exposure to valproic acid and autism--a current review of clinical and animal studies. Neurotoxicol. Teratol. 36, 47–56. doi:10.1016/j.ntt.2013.01.004

Roy, C.C., Kien, C.L., Bouthillier, L., Levy, E., 2006. Short-chain fatty acids: ready for prime time? Nutr. Clin. Pract. Off. Publ. Am. Soc. Parenter. Enter. Nutr. 21, 351–366.

Ruff, J.S., Nelson, A.C., Kubinak, J.L., Potts, W.K., 2012. MHC signaling during social communication. Adv. Exp. Med. Biol. 738, 290–313. doi:10.1007/978-1-4614-1680-7_17

Rumberger, J.M., Arch, J.R.S., Green, A., 2014. Butyrate and other short-chain fatty acids increase the rate of lipolysis in 3T3-L1 adipocytes. PeerJ 2. doi:10.7717/peerj.611

Ryu, H., Smith, K., Camelo, S.I., Carreras, I., Lee, J., Iglesias, A.H., Dangond, F., Cormier, K.A., Cudkowicz, M.E., Brown, R.H., Ferrante, R.J., 2005. Sodium phenylbutyrate prolongs survival and regulates expression of anti-apoptotic genes in transgenic amyotrophic lateral sclerosis mice. J. Neurochem. 93, 1087–1098. doi:10.1111/j.1471-4159.2005.03077.x

Sampson, T.R., Mazmanian, S.K., 2015. Control of brain development, function, and behavior by the microbiome. Cell Host Microbe 17, 565–576. doi:10.1016/j.chom.2015.04.011

Sanchis-Segura, C., Lopez-Atalaya, J.P., Barco, A., 2009. Selective boosting of transcriptional and behavioral responses to drugs of abuse by histone deacetylase inhibition. Neuropsychopharmacol. Off. Publ. Am. Coll. Neuropsychopharmacol. 34, 2642–2654. doi:10.1038/npp.2009.125

Santos, P.S.C., Schinemann, J.A., Gabardo, J., Bicalho, M. da G., 2005. New evidence that the MHC influences odor perception in humans: a study with 58 Southern Brazilian students. Horm. Behav. 47, 384–388. doi:10.1016/j.yhbeh.2004.11.005

Sarna, G.S., Bradbury, M.W., Cremer, J.E., Lai, J.C., Teal, H.M., 1979. Brain metabolism and specific transport at the blood-brain barrier after portocaval anastomosis in the rat. Brain Res. 160, 69–83.

Schellinck, H.M., Brown, R.E., Slotnick, B.M., 1991. Training rats to discriminate between the odors of individual conspecifics. Anim. Learn. Behav. 19, 223–233. doi:10.3758/BF03197880

Scheppach, W., 1996. Treatment of distal ulcerative colitis with short-chain fatty acid enemas. A placebo-controlled trial. German-Austrian SCFA Study Group. Dig. Dis. Sci. 41, 2254–2259.

Schroeder, F.A., Lin, C.L., Crusio, W.E., Akbarian, S., 2007. Antidepressant-like effects of the histone deacetylase inhibitor, sodium butyrate, in the mouse. Biol. Psychiatry 62, 55–64. doi:10.1016/j.biopsych.2006.06.036

Schroeder, F.A., Penta, K.L., Matevossian, A., Jones, S.R., Konradi, C., Tapper, A.R., Akbarian, S., 2008. Drug-induced activation of dopamine D(1) receptor signaling and inhibition of class I/II histone deacetylase induce chromatin remodeling in reward circuitry and modulate cocaine-related behaviors. Neuropsychopharmacol. Off. Publ. Am. Coll. Neuropsychopharmacol. 33, 2981–2992. doi:10.1038/npp.2008.15

Sealy, L., Chalkley, R., 1978. The effect of sodium butyrate on histone modification. Cell 14, 115–121. doi:10.1016/0092-8674(78)90306-9

Selkrig, J., Wong, P., Zhang, X., Pettersson, S., 2014. Metabolic tinkering by the gut microbiome: Implications for brain development and function. Gut Microbes 5, 0–1.

Sharma, S., Taliyan, R., Singh, S., 2015. Beneficial effects of sodium butyrate in 6-OHDA induced neurotoxicity and behavioral abnormalities: Modulation of histone deacetylase activity. Behav. Brain Res. 291, 306–314. doi:10.1016/j.bbr.2015.05.052

Shen, H.-Y., Kalda, A., Yu, L., Ferrara, J., Zhu, J., Chen, J.-F., 2008. Additive effects of histone deacetylase inhibitors and amphetamine on histone H4 acetylation, cAMP responsive element binding protein

phosphorylation and DeltaFosB expression in the striatum and locomotor sensitization in mice. Neuroscience 157, 644–655. doi:10.1016/j.neuroscience.2008.09.019

Shimazu, T., Hirschey, M.D., Huang, J.-Y., Ho, L.T.Y., Verdin, E., 2010. Acetate metabolism and aging: an emerging connection. Mech. Ageing Dev. doi:10.1016/j.mad.2010.05.001

Shimazu, T., Hirschey, M.D., Newman, J., He, W., Shirakawa, K., Le Moan, N., Grueter, C.A., Lim, H., Saunders, L.R., Stevens, R.D., Newgard, C.B., Farese, R.V., Jr, de Cabo, R., Ulrich, S., Akassoglou, K., Verdin, E., 2013. Suppression of oxidative stress by β-hydroxybutyrate, an endogenous histone deacetylase inhibitor. Science 339, 211–214. doi:10.1126/science.1227166

Shin, H.J., Anzai, N., Enomoto, A., He, X., Kim, D.K., Endou, H., Kanai, Y., 2007. Novel liver-specific organic anion transporter OAT7 that operates the exchange of sulfate conjugates for short chain fatty acid butyrate. Hepatol. Baltim. Md 45, 1046–1055. doi:10.1002/hep.21596

Siigur, U., Ormisson, A., Tamm, A., 1993. Faecal short-chain fatty acids in breast-fed and bottle-fed infants. Acta Paediatr. Oslo Nor. 1992 82, 536–538.

Singer, A.G., Beauchamp, G.K., Yamazaki, K., 1997. Volatile signals of the major histocompatibility complex in male mouse urine. Proc. Natl. Acad. Sci. U. S. A. 94, 2210–2214.

Singh, N., Gurav, A., Sivaprakasam, S., Brady, E., Padia, R., Shi, H., Thangaraju, M., Prasad, P.D., Manicassamy, S., Munn, D.H., Lee, J.R., Offermanns, S., Ganapathy, V., 2014. Activation of Gpr109a, receptor for niacin and the commensal metabolite butyrate, suppresses colonic inflammation and carcinogenesis. Immunity 40, 128–139. doi:10.1016/j.immuni.2013.12.007

Singh, P.B., Herbert, J., Roser, B., Arnott, L., Tucker, D.K., Brown, R.E., 1990. Rearing rats in a germ-free environment eliminates their odors of individuality. J. Chem. Ecol. 16, 1667–1682. doi:10.1007/BF01014099

Sleeth, M.L., Thompson, E.L., Ford, H.E., Zac-Varghese, S.E.K., Frost, G., 2010. Free fatty acid receptor 2 and nutrient sensing: a proposed role for fibre, fermentable carbohydrates and short-chain fatty acids in appetite regulation. Nutr. Res. Rev. 23, 135–145. doi:10.1017/S0954422410000089

Smith, P.M., Howitt, M.R., Panikov, N., Michaud, M., Gallini, C.A., Bohlooly-Y, M., Glickman, J.N., Garrett, W.S., 2013. The Microbial Metabolites, Short-Chain Fatty Acids, Regulate Colonic Treg Cell Homeostasis. Science 341, 569–573. doi:10.1126/science.1241165

Spange, S., Wagner, T., Heinzel, T., Krämer, O.H., 2009. Acetylation of non-histone proteins modulates cellular signalling at multiple levels. Int. J. Biochem. Cell Biol. 41, 185–198. doi:10.1016/j.biocel.2008.08.027

Stackebrandt, E., Kramer, I., Swiderski, J., Hippe, H., 1999. Phylogenetic basis for a taxonomic dissection of the genus Clostridium. FEMS Immunol. Med. Microbiol. 24, 253–258.

Stilling, R.M., Bordenstein, S.R., Dinan, T.G., Cryan, J.F., 2014a. Friends with social benefits: host-microbe interactions as a driver of brain evolution and development? Front. Cell. Infect. Microbiol. 4, 147. doi:10.3389/fcimb.2014.00147

Stilling, R.M., Dinan, T.G., Cryan, J.F., 2014b. Microbial genes, brain & behaviour - epigenetic regulation of the gut-brain axis. Genes Brain Behav. 13, 69–86. doi:10.1111/gbb.12109

Stilling, R.M., Fischer, A., 2011. The role of histone acetylation in age-associated memory impairment and Alzheimer's disease. Neurobiol. Learn. Mem. 96, 19–26. doi:10.1016/j.nlm.2011.04.002

Sun, J., Ling, Z., Wang, F., Chen, W., Li, H., Jin, J., Zhang, H., Pang, M., Yu, J., Liu, J., 2016. Clostridium butyricum pretreatment attenuates cerebral ischemia/reperfusion injury in mice via anti-oxidation and anti-apoptosis. Neurosci. Lett. 613, 30–35. doi:10.1016/j.neulet.2015.12.047

Suzuki, T., Yoshida, S., Hara, H., 2008. Physiological concentrations of short-chain fatty acids immediately suppress colonic epithelial permeability. Br. J. Nutr. 100, 297–305. doi:10.1017/S0007114508888733

Sweatt, J.D., 2013. The Emerging Field of Neuroepigenetics. Neuron 80, 624–632. doi:10.1016/j.neuron.2013.10.023

Taggart, A.K.P., Kero, J., Gan, X., Cai, T.-Q., Cheng, K., Ippolito, M., Ren, N., Kaplan, R., Wu, K., Wu, T.-J., Jin,

L., Liaw, C., Chen, R., Richman, J., Connolly, D., Offermanns, S., Wright, S.D., Waters, M.G., 2005. (D)-beta-Hydroxybutyrate inhibits adipocyte lipolysis via the nicotinic acid receptor PUMA-G. J. Biol. Chem. 280, 26649–26652. doi:10.1074/jbc.C500213200

Takahashi, K., Nishida, A., Fujimoto, T., Fujii, M., Shioya, M., Imaeda, H., Inatomi, O., Bamba, S., Andoh, A., Sugimoto, M., 2016. Reduced Abundance of Butyrate-Producing Bacteria Species in the Fecal Microbial Community in Crohn's Disease. Digestion 93, 59–65. doi:10.1159/000441768

Takuma, K., Hara, Y., Kataoka, S., Kawanai, T., Maeda, Y., Watanabe, R., Takano, E., Hayata-Takano, A., Hashimoto, H., Ago, Y., Matsuda, T., 2014. Chronic treatment with valproic acid or sodium butyrate attenuates novel object recognition deficits and hippocampal dendritic spine loss in a mouse model of autism. Pharmacol. Biochem. Behav. 126, 43–49. doi:10.1016/j.pbb.2014.08.013

Tang, H., Lu, J.Y.-L., Zheng, X., Yang, Y., Reagan, J.D., 2008. The psoriasis drug monomethylfumarate is a potent nicotinic acid receptor agonist. Biochem. Biophys. Res. Commun. 375, 562–565. doi:10.1016/j.bbrc.2008.08.041

Tarini, J., Wolever, T.M.S., 2010. The fermentable fibre inulin increases postprandial serum short-chain fatty acids and reduces free-fatty acids and ghrelin in healthy subjects. Appl. Physiol. Nutr. Metab. 35, 9–16. doi:10.1139/H09-119

Tazoe, H., Otomo, Y., Karaki, S.-I., Kato, I., Fukami, Y., Terasaki, M., Kuwahara, A., 2009. Expression of short-chain fatty acid receptor GPR41 in the human colon. Biomed. Res. Tokyo Jpn. 30, 149–156.

Thangaraju, M., Cresci, G.A., Liu, K., Ananth, S., Gnanaprakasam, J.P., Browning, D.D., Mellinger, J.D., Smith, S.B., Digby, G.J., Lambert, N.A., Prasad, P.D., Ganapathy, V., 2009. GPR109A is a G-protein-coupled receptor for the bacterial fermentation product butyrate and functions as a tumor suppressor in colon. Cancer Res. 69, 2826–2832. doi:10.1158/0008-5472.CAN-08-4466

Theis, K.R., Schmidt, T.M., Holekamp, K.E., 2012. Evidence for a bacterial mechanism for group-specific social odors among hyenas. Sci. Rep. 2. doi:10.1038/srep00615

Theis, K.R., Venkataraman, A., Dycus, J.A., Koonter, K.D., Schmitt-Matzen, E.N., Wagner, A.P., Holekamp, K.E., Schmidt, T.M., 2013. Symbiotic bacteria appear to mediate hyena social odors. Proc. Natl. Acad. Sci. 110, 19832–19837. doi:10.1073/pnas.1306477110

Thomas, R.H., Meeking, M.M., Mepham, J.R., Tichenoff, L., Possmayer, F., Liu, S., MacFabe, D.F., 2012. The enteric bacterial metabolite propionic acid alters brain and plasma phospholipid molecular species: further development of a rodent model of autism spectrum disorders. J. Neuroinflammation 9, 153. doi:10.1186/1742-2094-9-153

Titgemeyer, E.C., Mamedova, L.K., Spivey, K.S., Farney, J.K., Bradford, B.J., 2011. An unusual distribution of the niacin receptor in cattle. J. Dairy Sci. 94, 4962–4967. doi:10.3168/jds.2011-4193

Topping, D.L., Clifton, P.M., 2001. Short-Chain Fatty Acids and Human Colonic Function: Roles of Resistant Starch and Nonstarch Polysaccharides. Physiol. Rev. 81, 1031–1064.

Troyer, K., 1984. Microbes, herbivory and the evolution of social behavior. J. Theor. Biol. 106, 157–169.

Ulven, T., 2012. Short-chain free fatty acid receptors FFA2/GPR43 and FFA3/GPR41 as new potential therapeutic targets. Front. Endocrinol. 3, 111. doi:10.3389/fendo.2012.00111

Van den Abbeele, P., Belzer, C., Goossens, M., Kleerebezem, M., De Vos, W.M., Thas, O., De Weirdt, R., Kerckhof, F.-M., Van de Wiele, T., 2013. Butyrate-producing Clostridium cluster XIVa species specifically colonize mucins in an in vitro gut model. ISME J. 7, 949–961. doi:10.1038/ismej.2012.158

Vecsey, C.G., Hawk, J.D., Lattal, K.M., Stein, J.M., Fabian, S.A., Attner, M.A., Cabrera, S.M., McDonough, C.B., Brindle, P.K., Abel, T., Wood, M.A., 2007. Histone Deacetylase Inhibitors Enhance Memory and Synaptic Plasticity via CREB: CBP-Dependent Transcriptional Activation. J Neurosci 27, 6128–6140. doi:10.1523/JNEUROSCI.0296-07.2007

Veiga, P., Pons, N., Agrawal, A., Oozeer, R., Guyonnet, D., Brazeilles, R., Faurie, J.-M., van Hylckama Vlieg,

J.E.T., Houghton, L.A., Whorwell, P.J., Ehrlich, S.D., Kennedy, S.P., 2014. Changes of the human gut microbiome induced by a fermented milk product. Sci. Rep. 4, 6328. doi:10.1038/srep06328

Verbeke, K.A., Boobis, A.R., Chiodini, A., Edwards, C.A., Franck, A., Kleerebezem, M., Nauta, A., Raes, J., van Tol, E.A.F., Tuohy, K.M., 2015. Towards microbial fermentation metabolites as markers for health benefits of prebiotics. Nutr. Res. Rev. 28, 42–66. doi:10.1017/S0954422415000037

Vieira, E.L.M., Leonel, A.J., Sad, A.P., Beltrão, N.R.M., Costa, T.F., Ferreira, T.M.R., Gomes-Santos, A.C., Faria, A.M.C., Peluzio, M.C.G., Cara, D.C., Alvarez-Leite, J.I., 2012. Oral administration of sodium butyrate attenuates inflammation and mucosal lesion in experimental acute ulcerative colitis. J. Nutr. Biochem. 23, 430–436. doi:10.1016/j.jnutbio.2011.01.007

Vijay, N., Morris, M.E., 2014. Role of Monocarboxylate Transporters in Drug Delivery to the Brain. Curr. Pharm. Des. 20, 1487–1498.

Viosca, J., Jancic, D., López-Atalaya, J.P., Benito, E., 2007. Hunting for Synaptic Tagging and Capture in Memory Formation. J. Neurosci. 27, 12761–12763. doi:10.1523/JNEUROSCI.4093-07.2007

Vital, M., Howe, A.C., Tiedje, J.M., 2014. Revealing the bacterial butyrate synthesis pathways by analyzing (meta)genomic data. mBio 5, e00889. doi:10.1128/mBio.00889-14

Wakade, C., Chong, R., Bradley, E., Thomas, B., Morgan, J., 2014. Upregulation of GPR109A in Parkinson's disease. PloS One 9, e109818. doi:10.1371/journal.pone.0109818

Walker, A.W., Duncan, S.H., McWilliam Leitch, E.C., Child, M.W., Flint, H.J., 2005. pH and Peptide Supply Can Radically Alter Bacterial Populations and Short-Chain Fatty Acid Ratios within Microbial Communities from the Human Colon. Appl. Environ. Microbiol. 71, 3692–3700. doi:10.1128/AEM.71.7.3692-3700.2005

Wang, H.-B., Wang, P.-Y., Wang, X., Wan, Y.-L., Liu, Y.-C., 2012. Butyrate enhances intestinal epithelial barrier function via up-regulation of tight junction protein Claudin-1 transcription. Dig. Dis. Sci. 57, 3126–3135. doi:10.1007/s10620-012-2259-4

Wedekind, C., Seebeck, T., Bettens, F., Paepke, A.J., 1995. MHC-dependent mate preferences in humans. Proc. Biol. Sci. 260, 245–249. doi:10.1098/rspb.1995.0087

Wei, Y., Melas, P.A., Wegener, G., Mathé, A.A., Lavebratt, C., 2015. Antidepressant-like effect of sodium butyrate is associated with an increase in TET1 and in 5-hydroxymethylation levels in the Bdnf gene. Int. J. Neuropsychopharmacol. Off. Sci. J. Coll. Int. Neuropsychopharmacol. CINP 18. doi:10.1093/ijnp/pyu032

Won, Y.-J., Lu, V.B., Puhl, H.L., Ikeda, S.R., 2013. β-Hydroxybutyrate modulates N-type calcium channels in rat sympathetic neurons by acting as an agonist for the G-protein-coupled receptor FFA3. J. Neurosci. Off. J. Soc. Neurosci. 33, 19314–19325. doi:10.1523/JNEUROSCI.3102-13.2013

Yadav, H., Lee, J.-H., Lloyd, J., Walter, P., Rane, S.G., 2013. Beneficial Metabolic Effects of a Probiotic via Butyrate-induced GLP-1 Hormone Secretion. J. Biol. Chem. 288, 25088–25097. doi:10.1074/jbc.M113.452516

Yano, J.M., Yu, K., Donaldson, G.P., Shastri, G.G., Ann, P., Ma, L., Nagler, C.R., Ismagilov, R.F., Mazmanian, S.K., Hsiao, E.Y., 2015. Indigenous bacteria from the gut microbiota regulate host serotonin biosynthesis. Cell 161, 264–276. doi:10.1016/j.cell.2015.02.047

Yonezawa, T., Kurata, R., Yoshida, K., Murayama, M.A., Cui, X., Hasegawa, A., 2013. Free fatty acids-sensing G protein-coupled receptors in drug targeting and therapeutics. Curr. Med. Chem. 20, 3855–3871.

Yudkoff, M., Daikhin, Y., Nissim, I., Lazarow, A., Nissim, I., 2001. Brain amino acid metabolism and ketosis. J. Neurosci. Res. 66, 272–281.

Zarling, E.J., Ruchim, M.A., 1987. Protein origin of the volatile fatty acids isobutyrate and isovalerate in human stool. J. Lab. Clin. Med. 109, 566–570.

Zhao, S., Xu, W., Jiang, W., Yu, W., Lin, Y., Zhang, T., Yao, J., Zhou, L., Zeng, Y., Li, H., Li, Y., Shi, J., An, W., Hancock, S.M., He, F., Qin, L., Chin, J., Yang, P., Chen, X., Lei, Q., Xiong, Y., Guan, K.-L., 2010. Regulation of Cellular Metabolism by Protein Lysine Acetylation. Science 327, 1000–1004. doi:10.1126/science.1179689

Zheng, P., Zeng, B., Zhou, C., Liu, M., Fang, Z., Xu, X., Zeng, L., Chen, J., Fan, S., Du, X., Zhang, X., Yang, D.,

Yang, Y., Meng, H., Li, W., Melgiri, N.D., Licinio, J., Wei, H., Xie, P., 2016. Gut microbiome remodeling induces depressive-like behaviors through a pathway mediated by the host's metabolism. Mol. Psychiatry. doi:10.1038/mp.2016.44

Zimmerman, M.A., Singh, N., Martin, P.M., Thangaraju, M., Ganapathy, V., Waller, J.L., Shi, H., Robertson, K.D., Munn, D.H., Liu, K., 2012. Butyrate suppresses colonic inflammation through HDAC1-dependent Fas upregulation and Fas-mediated apoptosis of T cells. Am. J. Physiol. - Gastrointest. Liver Physiol. 302, G1405–G1415. doi:10.1152/ajpgi.00543.2011

Zou, X.-H., Li, H.-M., Wang, S., Leski, M., Yao, Y.-C., Yang, X.-D., Huang, Q.-J., Chen, G.-Q., 2009. The effect of 3-hydroxybutyrate methyl ester on learning and memory in mice. Biomaterials 30, 1532–1541. doi:10.1016/j.biomaterials.2008.12.012

14 Figure legends

Figure 1: (A) Structual representations of butyrate and related molecules. (B) Strongly simplified diagram of host-microbiota co-metabolism of butyrate. For more details see (Louis and Flint, 2009; Macfarlane and Macfarlane, 2003). Acetyl-CoA: acetyl coenzyme A; TCA: Tricarboxylic acid cycle (citric acid cycle/Krebs cycle)

Figure 2: **(A)** Histone acetylation contributes to the formation of open, active chromatin (euchromatin). Acetylation of the lysine (K) tails of the 4 nucleosomal histones (mainly histones H3 and H4) is catalysed by the opposing activity of histone/K-lysine acetyltransferases (HATs/KATs) and histone/K-lysine deacetylases (HDACs/KDACs). **(B)** Approximate IC50 values for different synthetic and natural or endogenous HDAC inhibitors, based on *in vitro* studies. Note that for pyruvate different values found can be found in the literature. TSA: Trichostatin A, SAHA: suberanilohydroxamic acid

Figure 3: Diagram depicting the events occurring in the synaptic tagging and capturing model and how this pathway can be "short-cut" by the use of HDAC inhibitors (HDACi) or HAT activators, both promoting histone acetylation and nuclear gene expression necessary for long-term memory.

Figure 4: Schematic summary of butyrate effects on host physiology and brain function

15 Table captions

Table 1: Cellular receptors for butyrate. Form: Formate, Ac: Acetate, Prop: Propionate, But: butyrate, Val: Valerate (Pentanoate, C5), Capr: Caproate (Hexanoate, C6). *Note that there is conflicting evidence for D-β-hydroxybutyrate (DHB) regarding agonist/antagonist status at FFAR3. For further synthetic ligands (agonists and antagonists) for FFAR2 and FFAR3 see (Ulven, 2012), for HCAR2 see http://www.uniprot.org/uniprot/Q8TDS4#function.

Table 2: Selected *in vivo* studies using butyrate, 4-phenylbutyrate or butyrate-producing bacteria to modulate brain physiology and function.

16 Boxes

Box 1: Dietary fibre is a rather unspecific term comprising all host-indigestible dietary carbohydrates, i.e. polysaccharides mainly found in plants and mammalian milk and dairy products (Topping and Clifton, 2001). They can be further subdivided by their solubility in water, their specific sugar monomer and/or polymerisation complexity. An important, well-studied class of soluble fibre is short (3 to 10 monomers) oligosaccharides made from fructose or galactose (FOS and GOS). These can be found for example in agave, bananas, onions and garlic. Other, glucose-based fibre classes are resistant starch (RS), found in e.g. cooled boiled potatoes, β-glucans, found in oat, barley, wheat, and rye, and cellulose, the main plant cell wall component. In addition, non-starch polymers of xylose and other sugars (xylans and other hemicelluloses) as well as uronic acids (pectins) are found in plant-based diets, most prominently in pears, apples, guavas, plums, and oranges (Bindelle et al., 2008). Prebiotics are defined as food supplements that specifically promote growth of health-associated bacteria in the gut. They also are usually non-digestible carbohydrates that reach the caecum to become substrates for microbial fermentation (Cummings et al., 2001; Topping and Clifton, 2001).