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Targeted nutrition to promote metabolic health – the untapped power of phytochemicals

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1. The global obesity epidemic

Obesity is a global problem of epidemic proportion and considered to be one of the top five threats to mankind according to a recent paper published by the WHO. Based on a study from the NIH obesity is thought to be responsible for 300'000 deaths worldwide each year and thus constitutes the second leading cause of preventable death after smoking.

Based on this global threat the first important question is how obesity can be defined. Normally obesity is characterized by an increase in body fat mass beyond the normal range. A way to express this on the population level is the use of the body mass index (BMI), which is calculated from the weight (in kg) divided by the squared height (in m). A BMI of more than 25 is considered as overweight, while a BMI of 30 or more is considered as obese with different gradings to indicate the degree of severity. For many years, there has been a lot of criticism of the BMI as for example people with higher muscle mass have an exaggerated BMI. Nevertheless, it should be noted that on a population level the BMI has been useful due to its simple calculation and its stable basis of measurements.[1]

While in most areas of the world the rate of obesity is increasing at an alarming rate Swiss obesity rates have remained relatively stable at a high level over the last years. The BAG currently estimates that approximately 42% of all Swiss adults are overweight, while approximately 11% can be considered obese (i.e. with a BMI of 30 or more.[2]

Based on the etiology of obesity one important question to ask is, why is obesity developing at such an alarming rate and what are the main factors driving the progression of obesity. The simple answer to this question is that weight gain which ultimately leads to obesity is due to a positive energy balance, *i.e.*, a person, often over long periods of time, takes in more calories than he or she actually consumes. A simple calculation is as follows: an extra cookie with an energy content of 60 kcal each day amounts to a weight gain of 7 g of fat per day. This would lead to a weight gain of 2.5 kg per year, which would be an extra 50 kg within 20 years. While this sounds like a trivial problem, which should be easy solvable the worldwide progression of obesity despite numerous interventions, educational programs, dietary strategies, etc. clearly indicates that living in a negative energy balance is difficult for most people. Nevertheless, most nutritional intervention strategies for obesity aim to elicit a negative energy balance with somewhat limited success.

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1 Seidell JC, Flegal KM. Assessing obesity: classification and epidemiology. Br Med Bull. 1997;53(2):238–52.

2 BAG B für G. Übergewicht und Adipositas [Internet]. [cited 2021 Feb 5]. Available from: *https://www.bag.admin.ch/bag/de/home/gesund-leben/gesundheitsfoerderung-und-praevention/koerpergewicht/uebergewicht-und-adipositas.html*

2. Obesity and metabolic health, a different view

Taken together, the points mentioned above trigger a further question. If obesity itself is so difficult to treat, would it be possible to treat the obesity associated problems which are the cause for the increased mortality and morbidity rates? To answer this question, it is important to have a closer look at the reasons for obesity linked mortality as obesity itself clearly is not a direct cause of death. First and foremost, obesity is linked to an increased risk for cardiovascular diseases (such as myocardial infarction and stroke), which is still the number one cause of death worldwide.[3] Secondly obesity predisposes people to develop type 2 diabetes, which is one of the most important drivers of health care costs in addition to severely impacting the quality of life as well as the life expectancy. Other reported co-morbidities are neurodegenerative disorders as well as certain types of cancers to name but a few, which similarly contribute to the increased rates of mortality observed in the obese population.[4] Taken together one can conclude that obesity causes an unhealthy metabolic phenotype, which in turn leads to development of co-morbidities, which either affect life quality or expectancy. Is this fact a leverage that could be used in to reduce the mortality burden of obesity? One possible answer is based on a phenomenon, which has been gaining attention in recent years and is called the concept of metabolically healthy obesity. It is well documented that a certain percentage of obese subjects never develop any co-morbidities as outlined above for so far unknown reasons.[5] While at the populations level the risk for cardiovascular diseases is still increased above those of a healthy lean individual the incident rates are much lower than in unhealthy obese subjects.[6] Given the aforementioned problems to achieve a negative energy balance to several studies have aimed at understanding what constitutes a healthy obesity and how such a state could be promoted.

The example of healthy obese subjects suggests that excessive fat mass or quantity *per se* is not a primary determinant for severe health outcomes. In fact, the distribution of adipose tissue and its quality influence the differentiation between healthy *vs* unhealthy obesity. Deposition of adipose tissue within the leg or subcutaneous area are linked to better metabolic health than adipose tissue built up in the abdominal cavity.[7] Similarly, lower amounts of lipids in organs not predisposed to lipid accumulation (e.g. liver, skeletal muscle) is metabolically favorable. Another factor which seem to be linked to metabolic health besides the anatomical location is adipose quality, which is determined by the tissue's architecture and functionality. One aspect that separates low from high quality adipose tissue is the degree of inflammation as well as the size of the individual adipocyte, however so far it is not fully clear if the latter two points are cause or consequence of healthy adipose tissue.[5]

Another aspect of adipose tissue quality which has received renewed attention in the last few years is a second, much smaller adipose organ formed by brown adipocytes. These highly specialized cells can burn energy from glucose or lipids as heat, thereby increasing energy expenditure and promoting metabolic flux. Newborn humans are equipped with high amounts of this thermogenic brown adipose tissue (BAT) to defend body temperature in cold.[8] However, BAT activity and volume decline with aging and obesity, leaving the tissue in a predominantly inert but activatable state. Cold acclimation, even in morbidly obese individuals, or pharmacological interventions with adrenergic receptor agonists can activate BAT function and stimulate BAT expansion.[9],[10] In this metabolically active state, BAT acts as nutrient sink to cover the elevated energy demand of the thermogenic process. An exciting question at this point is whether active BAT consumes enough calories to induce a negative energy balance able to trigger weight loss. With an estimated total BAT mass of 470–2200 g, the tissue would spend around 23–105 kcal/day at room temperature and up to 180 kcal/day when maximally activated by cold[11],[12] which would be sufficient to compensate for the "extra daily cookie" and more possibly induce a healthy met-

3 Wilson PWF, D'Agostino RB, Sullivan L, Parise H, Kannel WB. Overweight and obesity as determinants of cardiovascular risk: the Framingham experience. Arch Intern Med. 2002 Sep 9;162(16):1867–72.

4 González-Muniesa P, Mártinez-González M-A, Hu FB, Després J-P, Matsuzawa Y, Loos RJF, *et al.* Obesity. Nat Rev Dis Primers. 2017 Jun 15; 3:17034.

5 Blüher M. Metabolically Healthy Obesity. Endocr Rev. 2020 May 1;41(3).

6 Eckel N, Meidtner K, Kalle-Uhlmann T, Stefan N, Schulze MB. Metabolically healthy obesity and cardiovascular events: A systematic review and meta-analysis. Eur J Prev Cardiol. 2016 Jun;23(9):956–66.

7 Chen G-C, Arthur R, Iyengar NM, Kamensky V, Xue X, Wassertheil-Smoller S, *et al.* Association between regional body fat and cardiovascular disease risk among postmenopausal women with normal body mass index. Eur Heart J. 2019 Sep 7;40(34):2849–55.

8 Cannon B, Nedergaard J. Brown adipose tissue: function and physiological significance. Physiol Rev. 2004 Jan;84(1):277–359.

9 O'Mara AE, Johnson JW, Linderman JD, Brychta RJ, McGehee S, Fletcher LA, *et al.* Chronic mirabegron treatment increases human brown fat, HDL cholesterol, and insulin sensitivity. The Journal of Clinical Investigation. 2020 Jan.

10 Vijgen GHEJ, Bouvy ND, Teule GJJ, Brans B, Schrauwen P, van Marken Lichtenbelt WD. Brown adipose tissue in morbidly obese subjects. PloS One. 2011 Feb;6(2):e17247.

11 Carpentier AC, Blondin DP, Virtanen KA, Richard D, Haman F, Turcotte ÉE. Brown Adipose Tissue Energy Metabolism in Humans. Frontiers in Endocrinology [Internet]. 2018 Aug [cited 2019 Dec 5];9. Available from: *https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6090055/*

12 Leitner BP, Huang S, Brychta RJ, Duckworth CJ, Baskin AS, McGehee S, *et al.* Mapping of human brown adipose tissue in lean and obese young men. PNAS. 2017 Aug 8;114(32):8649–54.

abolic phenotype due to enhanced metabolic flux. The striking link between BAT and metabolic and cardiovascular health in humans was very recently demonstrated in a large retrospective study, which could show that individuals with BAT have improved metabolic health, a lower prevalence of type 2 diabetes, hypertension and abnormal blood lipid profiles.[13] Most importantly, this link is more prominent in obese compared to normal weight people, which suggests that although BAT activity is generally reduced with adiposity, obese people who maintain functional BAT are possibly protected against the adverse metabolic changes related to obesity.

How would one restore BAT? Reducing overnight temperature to 19°C is sufficient to stimulate BAT activity and enhance energy expenditure in lean men.[14] Yet, the insulating subcutaneous adipose tissue layer of obese subject overcomes this mild cold stimulus and more "extreme" cooling-protocols are unpleasant/impractical on a long run. Considering that overweight and obesity are affecting thousands of people, a broad approach without the need of prescriptive medicine is desirable. The success story of salt iodinisation in Switzerland to tackle iodine deficiency against goiter highlights the potential of targeted nutritional interventions to improve public health.[15] Therefore, could it be possible to apply this paradigm and exploit daily diet to activate brown fat by adding bioactive nutrients as supplements or by combining selected foods and thereby foster healthy obesity (see Figure 1)?

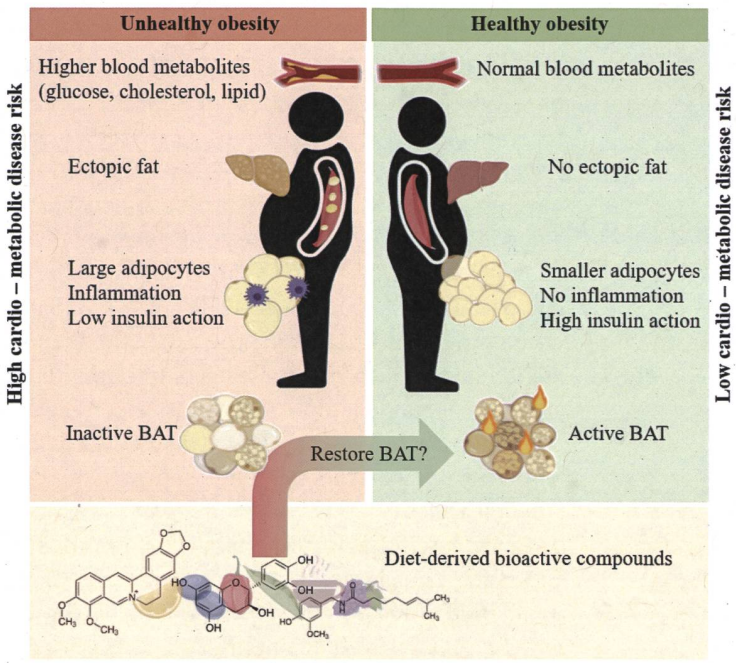

Figure 1. Conceptual illustration on how diet-derived bioactive compounds could promote the switch from unhealthy to healthy obesity by restoring functional competent brown adipose tissue.

3. Thermogenic foods, what is known?

A good example for a thermogenic food, based on the knowledge of the molecular components, are chili peppers, which elicit notable physiological reactions such as increased body temperature, vessel dilation and sweating as cooling-mechanism. The bioactive principles mediating these effects are the well-studied capsinoids. As already outlined, cold is the most potent activator of BAT thermogenesis. Sensory neurons in our body sense low temperatures by stimulating transient receptor potential (TRP) cation channels, which send an electric signal to the brain, more specifically to the hypothalamus. As response, the hypothalamus increases sympathetic nerve output (noradrenaline release), which activates the thermogenic brown adipocytes. Interestingly, sensory neurons are not only below the skin, but also within the gut, where ingested capsinoids are able to stimulate TRP channels and initiate the same physiological response as cold, thermogenesis.[16] It is thus little surprising that studies in rodents and humans report higher sympathetic nerve output after capsinoid intake.[17,18] Indeed, capsaicin acutely increases energy expenditure in humans within 2 hours of ingestion and this effect is dependent on brown adipose tissue activation.[19] Moreover, in people with low or absent BAT activity before the dietary intervention, daily capsinoid intake for 6 week enhances BAT activity as measured by an increase in cold-induced energy expenditure and lower body fat.[20] This underlines the potential of dietary components to activate BAT to a significant degree, even when BAT is initially not detectable as it is observed in metabolic unhealthy obese individuals.

13 Becher T, Palanisamy S, Kramer DJ, Eljalby M, Marx SJ, Wibmer AG, *et al.* Brown adipose tissue is associated with cardiometabolic health. Nat Med. 2021 Jan;27(1):58–65.

14 Chen KY, Brychta RJ, Linderman JD, Smith S, Courville A, Dieckmann W, *et al.* Brown fat activation mediates cold-induced thermogenesis in adult humans in response to a mild decrease in ambient temperature. J Clin Endocrinol Metab. 2013 Jul;98(7):E1218–1223.

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16 Bevan S, Szolcsányi J. Sensory neuron-specific actions of capsaicin: mechanisms and applications. Trends in Pharmacological Sciences. 1990 Aug;11(8):330–3.

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19 Yoneshiro T, Aita S, Kawai Y, Iwanaga T, Saito M. Nonpungent capsaicin analogs (capsinoids) increase energy expenditure through the activation of brown adipose tissue in humans. The American Journal of Clinical Nutrition. 2012 Apr;95(4):845–50.

20 Yoneshiro T, Aita S, Matsushita M, Kayahara T, Kameya T, Kawai Y, *et al.* Recruited brown adipose tissue as an antiobesity agent in humans. J Clin Invest. 2013 Aug;123(8):3404–8.

Another example which is derived based on epidemiological evidence rather than molecular evidence, are black and green teas which are the most consumed tea varieties globally. Epidemiological studies imply that high green-tea consumption is associated with lower blood pressure, lower fasting blood glucose and a reduced risk of cardiovascular diseases.[21,22,23] In addition to caffeine, catechins are the major bioactive compounds within green tea, which are also high in cocoa-products. Apart from the established antioxidant action of caffeine and catechins, both, human and animal studies imply that catechins strengthen metabolic health by increasing energy expenditure and stimulating BAT metabolism. Early data from isolated rat BAT show that catechins increase metabolic turnover. Interestingly, this effect is reinforced when BAT is additionally exposed to ephedrine (a sympathomimetic).[24] This potentiation of the activating adrenergic signals is a combination of two observations. First, catechins inhibit *in vitro* the noradrenaline-degrading enzyme, which could ensure a more long-lasting sympathetic stimulation of brown adipocytes.[25] Second, as the capsinoids, catechins can stimulate directly TRP channels, leading to sympathetic nerve activation.[26] In rats, catechin intake leads to a higher BAT mass,[27] and a comparable increase in BAT-density is observed young woman with daily catechin intake.[28] A single intake of a catechin-containing beverage can increase energy expenditure in healthy men with metabolically active BAT but is ineffective in men with no detectable BAT-activity. Remarkably, chronic catechin intake in these no-BAT men can recruit BAT and enhances cold-triggered thermogenesis as well as fat oxidation compared to the placebo-control group.[29] This promotion of BAT-activity was not accompanied by improved metabolic parameters. However, it is questionable to what extent an already "healthy" study subject could become "healthier". In obese or overweight people, catechin intake decreases cholesterol levels and multiple studies show that high catechin intake lowers BMI and waist circumference, thereby assisting weight loss and maintenance.[30,31,32]

Another promising molecule is berberine (BBR), which is not frequent in edible leaves, vegetable or fruits but can be isolated from the plants of the *berberis* genus for supplements. In various genetic or diet-induced rodent models of obesity, BBR-administration through the diet lowered weight gain and fat accumulation compared to control-fed animals. BBR-feeding further helped to maintain body temperature in mice during a cold-challenge and enhanced metabolic rate, which was accompanied by a healthier blood profile.[33] Most importantly, BBR-stimulated directly BAT-mediated thermogenesis and upregulated molecular markers of BAT activity (e.g. UCP1 protein). *In vitro* experiments further suggest that BRR boosts the formation of new brown adipocyte from precursor cells. First human studies investigating the effects of BBR intake on BAT activation and metabolic health provide encouraging results. In overweight subject, BBR intake for one month enhanced BAT activity and mass when compared in a before-after analysis.[33] Moreover, the percentage change in BAT activity positively correlated with weight change. Other studies with diabetic patients could show that, apart from weight loss, BBR improves blood glucose control and improves the body's sensitivity to insulin.[34] Whether or not BBR increases energy expenditure in humans is yet unknown but the available data implies that BRR

21 Xu R, Bai Y, Yang K, Chen G. Effects of green tea consumption on glycemic control: a systematic review and meta-analysis of randomized controlled trials. Nutrition & Metabolism. 2020 Jul 10;17(1):56.

22 Xu R, Yang K, Ding J, Chen G. Effect of green tea supplementation on blood pressure. Medicine (Baltimore) [Internet]. 2020 Feb 7 [cited 2021 Feb 5];99(6). Available from: *https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7015560/*

23 Nagao T, Hase T, Tokimitsu I. A Green Tea Extract High in Catechins Reduces Body Fat and Cardiovascular Risks in Humans. Obesity. 2007;15(6):1473–83.

24 Dulloo AG, Seydoux J, Girardier L, Chantre P, Vandermander J. Green tea and thermogenesis: interactions between catechin-polyphenols, caffeine and sympathetic activity. International Journal of Obesity and Related Metabolic Disorders: Journal of the International Association for the Study of Obesity. 2000 Feb;24(2):252–8.

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26 Kurogi M, Kawai Y, Nagatomo K, Tateyama M, Kubo Y, Saitoh O. Auto-oxidation products of epigallocatechin gallate activate TRPA1 and TRPV1 in sensory neurons. Chemical Senses. 2015 Jan;40(1):27–46.

27 Choo JJ. Green tea reduces body fat accretion caused by high-fat diet in rats through beta-adrenoceptor activation of thermogenesis in brown adipose tissue. The Journal of Nutritional Biochemistry. 2003 Nov;14(11):671–6.

28 Choo JJ. Green tea reduces body fat accretion caused by high-fat diet in rats through beta-adrenoceptor activation of thermogenesis in brown adipose tissue. The Journal of Nutritional Biochemistry. 2003 Nov;14(11):671–6.

29 Yoneshiro T, Matsushita M, Hibi M, Tone H, Takeshita M, Yasunaga K, *et al.* Tea catechin and caffeine activate brown adipose tissue and increase cold-induced thermogenic capacity in humans. The American Journal of Clinical Nutrition. 2017;105(4):873–81.

30 Auvichayapat P, Prapochanung M, Tunkamnerdthai O, Sripanidkulchai B, Auvichayapat N, Thinkhamrop B, *et al.* Effectiveness of green tea on weight reduction in obese Thais: A randomized, controlled trial. Physiology & Behavior. 2008 Feb;93(3):486–91.

31 Matsuyama T, Tanaka Y, Kamimaki I, Nagao T, Tokimitsu I. Catechin safely improved higher levels of fatness, blood pressure, and cholesterol in children. Obesity (Silver Spring, Md). 2008 Jun;16(6):1338–48.

32 Phung OJ, Baker WL, Matthews LJ, Lanosa M, Thorne A, Coleman CI. Effect of green tea catechins with or without caffeine on anthropometric measures: a systematic review and meta-analysis. The American Journal of Clinical Nutrition. 2010 Jan;91(1):73–81.

33 Wu L, Xia M, Duan Y, Zhang L, Jiang H, Hu X, *et al.* Berberine promotes the recruitment and activation of brown adipose tissue in mice and humans. Cell Death & Disease. 2019 Jun;10(6):468.

34 Yin J, Xing H, Ye J. Efficacy of berberine in patients with type 2 diabetes mellitus. Metabolism: Clinical and Experimental. 2008 May;57(5):712–7.

supplementation stimulates BAT function in humans and improves metabolic health.

Lastly, the phytochemicals present in the popular blueberries can modulate BAT activity, at least in mouse models. The blueberry anthocyanins are not only responsible for its color but also mediate some of its beneficial effect. Cyanidine-3-glucoside (C3G) improves obesity-associated metabolic complications in genetic and diet-induce mouse models by enhancing the mitochondrial function of BAT, which is reflected in a bigger density of mitochondria and higher abundancy of UCP1 protein. C3G-enriched diet helps to maintain normal glucose control and drastically enhances energy expenditure due to an increase in the thermogenic capacity of BAT.[35],[36] So far, data on C3G efficacy are limited to animal studies and future trials might reveal whether these favorable effects are reproducible in humans.

Based on these examples we can draw multiple conclusions. Firstly, there is much more data available on the effects of phytochemicals on BAT activation from animal studies. Active human BAT in adults was only discovered 12 years ago and the methods needed to determine human BAT-activity are very cumbersome.[37],[38] Nevertheless, the examples from BBR, catechins or capsinoids illustrate the potential of dietary or plant-derived compounds to activate human BAT and assist BAT restoration. To date, most of the trials investigating the impact of phytochemicals on human BAT are conducted in healthy and mostly Asian subjects. Based on genetic variation, there are differences in the metabolization of many compounds between Asian and Western people, which could affect the activity of the substances and eventually their physiological effect. Therefore, more trials in Western individuals are needed to confirm positive effects observed in Asians. Likewise, more trials including obese participants will resolve whether the potency of these phytochemicals is sufficient to regain functional BAT in obesity, when baseline BAT activity is low or inexistent. Lastly, the discussed compounds exemplify that different phytochemical act by distinct mechanisms. Some stimulate the sympathetic nervous system, thereby imitating cold. Others enhance the molecular features, which define a brown adipocyte (e.g. more UPC1 protein), while a third type increases BAT mass by stimulating the formation of brown adipocytes. This diversity could be leveraged by smartly combining food products with different mode of actions that ultimately converge in the same outcome: maximized efforts to restore BAT function.

4. How do we identify new healthy foods using the recent advances in analytics?

At this point, the question arises as how do we know which food or plants are of interest and which active compounds do they contain? The green tea example emphasizes that epidemiological evidence gained from data comparing disease prevalence or metabolic factors between population groups with high or low intake of certain foods deliver valuable hints on possible candidates. The efficacy of BBR further underlines that also non-edible plant parts are an immense pool containing potential substances which could be used as supplements. Accordingly, "traditional" evidence from herbal preparations and knowledge on their use against disease symptoms are an important resource. Vice versa, molecular evidence, as it was available for the red chili, can help us understand, why a certain food exerts an observable effect. From these considerations it becomes clear that the missing link between epidemiological evidence and molecular evidence are comprehensive analytical methods to decipher the chemical composition of a complex mixture for defined studies on their efficacy. In other terms, once a candidate is selected, a thorough understanding of the chemical composition is necessary to decipher the mode of action at the molecular level.[39] For a plant, the separation and detection of its chemical constituents are the first required steps toward determining the chemical composition. The process of formal identification consists of isolating pure constituent and identifying it nowadays mainly with ^{1}H and ^{13}C-Nuclear Magnetic Resonance (NMR). Plants of interest are first tested on suitable bioassays and initial hits further fractionated to narrow down selections of molecules until it is possible to isolation the active ingredient(s).[40] To improve this somewhat tedious procedure and to prevent the re-discovery of already known molecules, the

35 You Y, Han X, Guo J, Guo Y, Yin M, Liu G, *et al.* Cyanidin-3-glucoside attenuates high-fat and high-fructose diet-induced obesity by promoting the thermogenic capacity of brown adipose tissue. Journal of Functional Foods. 2018 Feb 1;41:62–71.

36 You Y, Yuan X, Liu X, Liang C, Meng M, Huang Y, *et al.* Cyanidin-3-glucoside increases whole body energy metabolism by upregulating brown adipose tissue mitochondrial function. Mol Nutr Food Res. 2017 Nov;61(11).

37 van Marken Lichtenbelt WD, Vanhommerig JW, Smulders NM, Drossaerts JMAFL, Kemerink GJ, Bouvy ND, *et al.* Cold-activated brown adipose tissue in healthy men. The New England Journal of Medicine. 2009 Apr;360(15):1500–8.

38 Cypess AM, Lehman S, Williams G, Tal I, Rodman D, Goldfine AB, *et al.* Identification and importance of brown adipose tissue in adult humans. The New England Journal of Medicine. 2009 Apr;360(15):1509–17.

39 Hou J, Zhang J, Yao C, Bauer R, Khan I, Wu W, *et al.* Deeper Chemical Perceptions for Better Traditional Chinese Medicine Standards. 2019;

40 Sumner LW, Amberg A, Barrett D, Beale MH, Beger R, Daykin CA, *et al.* Proposed minimum reporting standards for chemical analysis Chemical Analysis Working Group (CAWG) Metabolomics Standards Initiative (MSI). Metabolomics. 2007 Sep;3(3):211–21.

concept of dereplication emerged.[41,42] Dereplication or simply annotation, refers to a putative identification of compounds within a plant, whose certainty is assigned by different levels, which range from (1) identified the compound *via* (2) putatively annotated a compound within a plant to (3) identified similar compound classes within a plant to (4) unidentified compounds.[40] Thus, dereplication approaches are used to prioritize extracts and/or constituents before any isolation process and to eliminate known active substances at an early stage.

This type of putative identification is mainly performed on data obtained by Liquid-chromatography coupled to mass spectrometry or shortly referred to as LC-MS. All constituents identified within a given analysis of a plant constitute the metabolome, and the observation of the metabolome by LC-MS is named metabolite profiling.[43] Today, the most advanced techniques are ultrahigh pressure liquid chromatography (UHPLC) coupled with high-resolution mass spectrometry (HRMS), which generate metabolite profiling by combining proper chromatographic resolution with sensitive detection and high mass resolution power. In these analyses, the chemical information can be further combined with biological data from bioassays in a process called biochemometrics, which enables the discovery of markers related to a certain bioactivity.[44] The advantage of this approach is that also constituents present in small amounts can be recognized in crude extracts or fractions, especially if multiple plants containing the same active compound are tested.

To properly annotate compounds which are identified by biochemometrics in a first step all possible molecular formulas are calculated from the peaks from the HRMS data using also the information on the retention time within pre-existing data. A major step forward in the annotation process has been achieved with the emergence of molecular networks. Molecular networking is a visual representation of the relatedness of molecules and aims to organize similar fragmentation spectra (HRMS/MS) into clusters, considering that structurally similar molecules share spectral similarity.[43] Spectral similarity is assessed in term of the number of common fragments shared by two HRMS/MS spectra and the relative intensities of these common fragments. In such a visualization, each HRMS/MS spectrum is represented as a node, which is connected to similar spectra by an edge representing the level of similarity. Today the UHPLC-HRMS data processing workflow includes the ability to search databases to propose putative identifications against HRMS and HRMS/MS data and databases, which compile the majority of the published molecules are now available and can be consulted in terms of molecular formula and chemotaxonomy.[45] Combining biochemometric and molecular networking approaches was recently proposed to identify active components within food that could be used to design a targeted nutrition to promote health,[46] and such approaches seem to be a promising avenue to accelerate the discovery of active molecules.

Returning to the concept above, the thermogenic capacity of BAT can be reinforced by different cellular signaling pathways, which are sensitive to various molecules. Analytical approaches are thus indispensable to examine the effects of a food at the molecular level and to elucidate the mode of action of identified compounds in suitable bioassays. Consequently, this interdisciplinary work enables an evidence-based decision on which substances/food to use or even combine to achieve the highest stimulatory input and thereby improve metabolic health. ■

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