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A B S T R A C T
Epidemiological studies highlight the negative correlation between vitamin D levels and the incidence of many non-skeletal diseases including inflammatory diseases, cancer, and metabolic and neurological disorders. However, most randomized controlled trials (RCTs) with oral vitamin D supplementation give mixed results orare inconclusive. It has been said that “discovery commences with the awareness of anomaly”. The “anomaly” between our preclinical and clinical data provides the opportunity to propose an alternative paradigm to the vitamin D endocrine system: the vitamin D autacoid paradigm. In the vitamin D autacoid paradigm, the extraskeletal effects of vitamin D depend on the tissue reserves of vitamin D metabolites. These vitamin D autacoid systems are inducible oscillatory ecosystems in which 1,25D is produced, acts and is inactivated locally. In the vitamin D autacoid paradigm, attaining adequacy of vitamin D in the systemic circulation is necessary but not sufficient; we must also ensure the repletion of the tissue stores. The co-existence of two different vitamin D systems, endocrine and autacoid, with different functions and regulations leads to “significant shifts in the criteria determining the legitimacy both of problems and of proposed solutions”. With respect to our clinical trials of vitamin D supplementation for unconventional effects, the proposed solution is administering and quantifying vitamin D metabolites directly to the target tissue

Introduction

Epidemiological studies highlight the negative correlation between vitamin D levels and the incidence of many non-skeletal diseases including inflammatory diseases, cancer, and metabolic and neurological disorders [1]. Even if the association does not imply causation, a large number of laboratory experiments further support these extra-skeletal functions of vitamin D. Nevertheless, most randomized controlled trials (RCTs) with oral vitamin D supplementation on these such so-called unconventional health effects give mixed results or are inconclusive [2–14]. The possible reasons why the results of randomized controlled trials do not meet our expectancy have been already extensively discussed. Co-variation, reverse causality, long-term outcomes, population heterogeneity with individual differences in response to supplementation, and the issue of co-nutrient status are some reasons proposed (see for example [15–18]). Another more drastic possibility is that the paradigm in which we conduct our trials, namely, the vitamin D endocrine system, is not adequate to handle the non-skeletal effects. “Discovery commences with the awareness of anomaly” [19]. The discrepancy between our preclinical and clinical data should be considered as an opportune worth investigating anomaly.

The vitamin D endocrine system

The vitamin D endocrine system paradigm states that vitamin D, either produced in the skin upon UVB exposure or provided by the diet, circulates in the blood and is hydroxylated in the liver to form 25-hydroxyvitamin D (25D), and thereafter in the kidney to generate the most active vitamin D metabolite, namely, 1,25 dihydroxyvitamin D (1,25D). The blood concentration of 1,25D is strictly regulated and does not significantly change with vitamin D intake, except in the case of extreme vitamin D deficiency or vitamin D overload [20]. On the other hand, the blood level of 25D varies depending on vitamin D synthesis by the skin, dietary intake and vitamin D supplementation [20]. For example, 25D levels vary with sun exposure due to vitamin D skin synthesis, while circulating 1,25D levels remain nearly constant throughout the year [21,22]. Similarly, in adults, the daily ingestion of 1000 IU vitamin D3 increases the serum concentration of 25D, but the level of 1,25D does not change [23]. This mechanism is why our vitamin D clinical trials are based on 25D status and not 1,25D status. Note, however, that a meta-analysis of randomized controlled trials concluded that vitamin D supplementation could increase circulating 1,25D concentrations, but did not sufficiently affect calcium homeostasis [24]. Hence, even if vitamin D supplementation might enhance circulating 1,25D levels during some clinical trials, it is still not sufficient to affect the reference marker of the vitamin D endocrine system, which is calcium homeostasis. The physiology of the vitamin D endocrine system in which 1,25D blood levels are strictly regulated generates the paradox of our ongoing clinical trials of oral vitamin D supplementation; our oral vitamin D supplementation effectively increases circulating 25D, but the results provide no evidence of a functional effect on the reference markers that are serum 1,25D and calcium levels. In the absence of any positive control for the functional efficiency of vitamin D supplementation, these trials cannot be conclusive. In the endocrine paradigm, and in the absence of vitamin D deficiency, blood 1,25D is so tightly regulated that increasing its concentration by oral vitamin D supplementation to achieve unconventional effects without causing hypercalcaemia in trial participants is similar to trying to square the circle. In other words, we cannot handle the unconventional effects of vitamin D supplementation through the endocrine paradigm. For extra skeletal diseases, attaining vitamin D adequacy in the systemic circulation is necessary but not sufficient: we need a paradigm shift.

The vitamin D autacoid paradigm

The word “autacoid” comes from the Greek autos (self) and akos (remedy). In autacoid systems, molecules are produced on demand and act locally in the same cells or tissues through autocrine and paracrine signalling [25]. Some examples of autacoids are histamine, serotonin, bradykinin and several lipids involved in the modulation of inflammation, including members of the Specialized Pro-resolving Mediators (SPM) family [25,26]. Recently it has become evident that extra-liver and extra-renal metabolism of vitamin D does occur [27]. For example, cutaneous [28,29], fat [30], immune and nervous tissues [31,32], among others, can express the genes encoding the enzymes that activate vitamin D or 25D in 1,25D. The expression in these tissues of these enzymes together with the presence of the Vitamin D receptor (VDR) represents vitamin D autacoid systems [33–35]. Hence, when investigating the unconventional effects of vitamin D, we must address two frameworks: the endocrine and the autacoid paradigms. Several critical points differentiate the vitamin D autacoid systems from the endocrine system. They are inducible, for example by inflammatory

stimuli, and the local increase in 1,25D is transient and resolves itself by the induction of CYP24A1, which encodes the enzyme that inactivates 1,25D into 1,24,25D. This mechanism means that 1,25D is produced, acts and is inactivated locally. Autacoid systems are oscillating ecosystems that do not affect serum 1,25D. A corollary of the existence of such inducible vitamin D autacoid systems is that they are not constitutively turned on by the circulating levels of 1,25D. This mechanism makes sense if we consider, for example, that the immunomodulatory function of 1,25D must be limited both in time and in space at the foci of inflammation. In the autacoid framework, the regulation is achieved by inducing locally the production of 1,25D and the expression of the VDR.

Another distinctive point is that the local synthesis of 1,25D requires that its precursors, namely, vitamin D and 25D, have sufficient local bioavailability. It is commonly accepted that circulating 25D makes the bioavailable precursor pool for 1,25D. However, what is correct in the vitamin D endocrine framework may prove to be incomplete in the autacoid paradigm. In the vitamin D autacoid paradigm, not only the circulating 25D but also the tissue reserves of vitamin D metabolites are important for the non-conventional effects of vitamin D [36]. Hence, in the autacoid framework, it is necessary but not sufficient to attain adequacy of vitamin D in the systemic circulation; we also need vitamin D adequacy in the sites of storage [36]. Adipose, skin and muscle tissues are the main body stores [30,37]. In the vitamin D autacoid framework, these reserves are used to produce 1,25D in the microenvironment thereby controlling, for example, the resolution of inflammation in a restricted location. In addition to adipose, skin and muscle tissues, other sites of storage likely exist. For example, in the human brain, the highest levels of 25 D are found in the corpus callosum that does not contain detectable 1.25 D [38]. This observation suggests that 25D might be stored in the myelin sheath (Fig. 1A). If this hypothesis is correct, the degradation of the myelin sheath by microglial cells during demyelinating or neurodegenerative processes [39] makes possible the synthesis by microglial

cells of 1,25D from the myelin 25D stores [40]. This would in turn trigger the immunomodulatory and neuroprotective response mediated by 1,25D (Fig. 1B). Note that the local levels of 25D and 1,25D would be much higher in the inflammatory microenvironment than in the circulating blood or cerebrospinal fluid. In humans, 86% of the final volume of myelin is observed at 5 years, and reaches its maximum at 17 years [41]. This raises the question to know if an oral vitamin D supplementation in the adulthood is sufficient to replete the 25D stores outside of these critical periods and how long it will take. Myelination is also dynamic in adults and increases in response to external stimuli such as a learning stimulation. The possibility that the 25D storage capacity and therefore the response to oral vitamin D supplementation also depends on adult myelin plasticity warrants further investigation. Another point raised by the existence of autacoid vitamin D systems is the question of whether the local delivery of vitamin D or 25D directly at the tissue level would be a better option than the oral supplementation to replete the tissues stores and to achieve the unconventional effects of 1,25D without causing hypercalcaemia [36]. For example, could the transcutaneous delivery of vitamin D or 25D in the breast adipose tissue be more effective than oral supplementation on breast cancer incidence, progression or therapeutic response? Note that covering the female breast is specific to humans, recent in evolution and prevents the synthesis of vitamin D by the breast skin. We should perhaps evaluate whether some of the vitamin D and 25D that should have been locally produced by the skin naturally exposed to the sun is intended to be directly stored in the underlying adipose tissue as part of an evolutionary preventive mechanism against breast cancer.

Concluding remarks and perspectives

The unconventional effects of vitamin D cannot be fully accounted for by means of the endocrine paradigm used to address calcium homeostasis and rickets. Nearly a century after the discovery that rickets is an endocrine disease associated with low 25D blood levels, we are discovering that autacoid diseases associated with low vitamin D or 25D tissue levels also exist. However, this issue cannot be adequately handled within the framework of the vitamin D endocrine paradigm. The goal to attain adequacy of vitamin D in the systemic circulation is necessary for endocrine functions, but is insufficient for autacoid systems that also depend of the local reserve of vitamin D metabolites at the tissue level. Assuming the co-existence of two different vitamin D systems, endocrine and autacoid, with different functions and regulations leads to "significant shifts in the criteria determining the legitimacy both of problems and of proposed solutions" [19].

With respect to our clinical trials of vitamin D supplementation for unconventional effects, the proposed solution is administering and quantifying vitamin D metabolites directly to the target tissue.

Declaration of Competing Interest

The authors declare no conflict of interest.

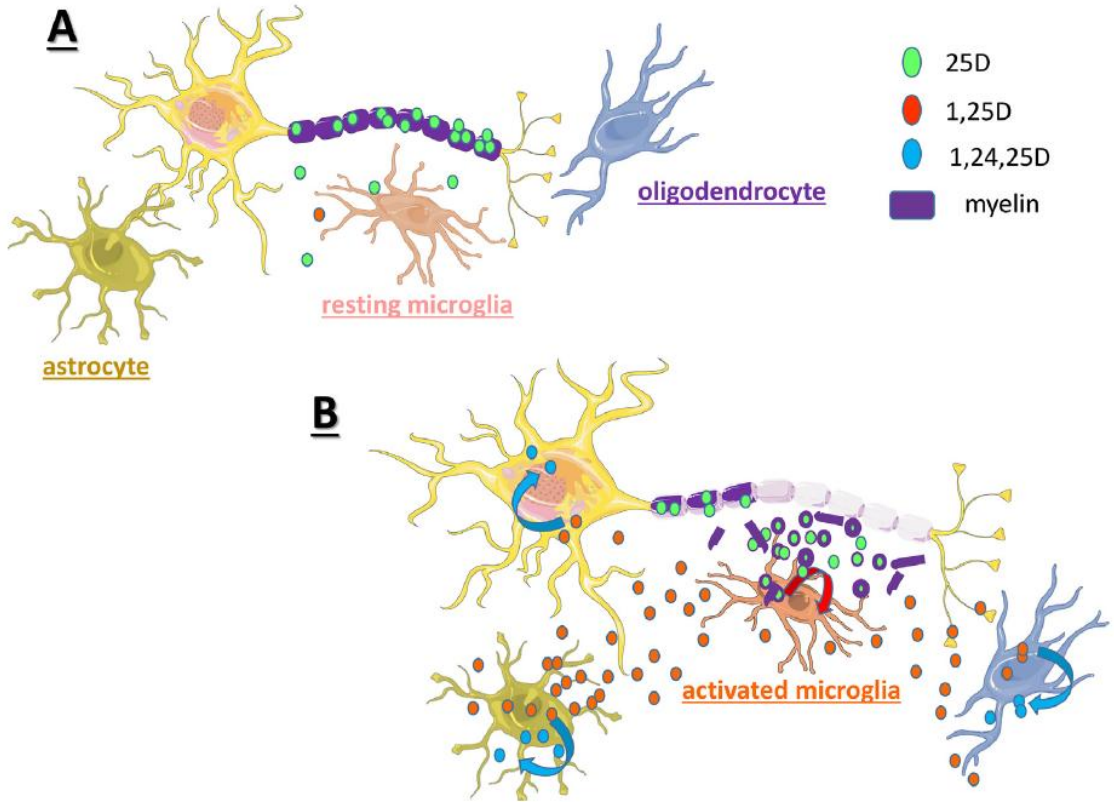

Fig. 1. In the human brain, the highest level of 25D is found in the myelin-rich corpus callosum. The storage of 25D in the myelin sheath makes sense from physicochemical reasons; 25D is lipophilic (A). Microglial cells actively participate in the clearance of myelin

debris produced either in the course of myelin turnover or during neuroinflammatory or neurodegenerative diseases [41]. Activated microglial cells also metabolizes 25D into 1,25D. In the proposed hypothetical autacoid model (B), the 25D myelin reserves are made available at the site of neurodegenerative processes because of myelin destruction. In the presence of myelin debris microglial cells metabolize 25D to 1,25D in the immediate extracellular space that would in turn trigger the immunomodulatory and neuroprotective function of 1,25D [31,42–44]. Note that when the autacoid vitamin D system is turned on, the level of 1,25D only increases in the microenvironment where it is much higher than its physiological blood or cerebrospinal fluid concentration. In autacoid systems, 1,25D is produced from the local tissue reserves, and it acts and is degraded locally. All these processes are independent of the vitamin D endocrine system [36].

References

[1] Holick MF. Vitamin D deficiency. N Engl J Med 2007;357:266–81. https://doi.org/10.1056/NEJMra070553.
[2] Autier P, Mullie P, Macacu A, Dragomir M, Boniol M, Coppens K, et al. Effect of vitamin D supplementation on non-skeletal disorders: a systematic review of metaanalyses and randomised trials. Lancet Diabetes Endocrinol 2017;5:986–1004. https://doi.org/10.1016/S2213-8587(17)30357-1.
[3] Giustina A, Adler RA, Binkley N, Bouillon R, Ebeling PR, Lazaretti-Castro M, et al. Controversies in vitamin D: summary statement from an international conference. J Clin Endocrinol Metab 2019;104:234–40. https://doi.org/10.1210/jc.2018-01414.
[4] Manson JE, Cook NR, Lee I-M, Christen W, Bassuk SS, Mora S, et al. Vitamin D supplements and prevention of cancer and cardiovascular disease. N Engl J Med 2019;380:33–44. https://doi.org/10.1056/NEJMoa1809944.
[5] Ng K, Nimeiri HS, McCleary NJ, Abrams TA, Yurgelun MB, Cleary JM, et al. Effect of high-dose vs standard-dose vitamin D3 supplementation on progression-free survival among patients with advanced or metastatic colorectal cancer: the sunshine randomized clinical trial. JAMA 2019;321:1370–9. https://doi.org/10.1001/jama.2019.2402.
[6] Gugger A, Marzel A, Orav EJ, Willett WC, Dawson-Hughes B, Theiler R, et al. Effect of monthly high-dose vitamin D on mental health in older adults: secondary analysis of a RCT. J Am Geriatr Soc 2019;67:1211–7. https://doi.org/10.1111/jgs.15808.
[7] Silvis K, Aronsson CA, Liu X, Uusitalo U, Yang J, Tamura R, et al. Maternal dietary supplement use and development of islet autoimmunity in the offspring: TEDDY study. Pediatr Diabetes 2019;20:86–92. https://doi.org/10.1111/pedi.12794.
[8] Bizzarri C, Pitocco D, Napoli N, Di Stasio E, Maggi D, Manfrini S, et al. No protective effect of calcitriol on beta-cell function in recent-onset type 1 diabetes: the IMDIAB XIII trial. Diabetes Care 2010;33:1962–3. https://doi.org/10.2337/dc10-0814.

[9] Quyyumi AA, Al Mheid I. The demise of vitamin D for cardiovascular prevention. JAMA Cardiol 2019. https://doi.org/10.1001/jamacardio.2019.1906.
[10] Barry EL, Passarelli MN, Baron JA. Vitamin D as cancer therapy?: Insights from 2 new trials. JAMA 2019;321:1354–5. https://doi.org/10.1001/jama.2019.2589.
[11] McGrath JJ. Vitamin D and mental health – the scrutiny of science delivers a sober message. Acta Psychiatr Scand 2017;135:183–4. https://doi.org/10.1111/acps.12708.

[12] Jorde R, Kubiak J. No improvement in depressive symptoms by vitamin D supplementation:
results from a randomised controlled trial. J Nutr Sci 2018;7:e30https://doi.org/10.1017/jns.2018.19.
[13] Maddock J, Zhou A, Cavadino A, Kuźma E, Bao Y, Smart MC, et al. Vitamin D and cognitive function: a Mendelian randomisation study. Sci Rep 2017;7:13230. https://doi.org/10.1038/s41598-017-13189-3.
[14] Rejnmark L, Bislev LS, Cashman KD, Eiríksdottir G, Gaksch M, Grübler M, et al. Non-skeletal health effects of vitamin D supplementation: a systematic review on findings from meta-analyses summarizing trial data. PLoS One 2017;12:e0180512https://doi.org/10.1371/journal.pone.0180512.
[15] Lappe JM, Heaney RP. Why randomized controlled trials of calcium and vitamin D sometimes fail. Dermato Endocrinol 2012;4:95–100. https://doi.org/10.4161/derm.19833.
[16] Grant WB, Boucher BJ, Bhattoa HP, Lahore H. Why vitamin D clinical trials should be based on 25-hydroxyvitamin D concentrations. J Steroid Biochem Mol Biol 2018;177:266–9. https://doi.org/10.1016/j.jsbmb.2017.08.009.
[17] Jorde R. RCTS are the only appropriate way to demonstrate the role of vitamin D in health. J Steroid Biochem Mol Biol 2018;177:10–4. https://doi.org/10.1016/j.jsbmb.2017.05.004.
[18] Scragg R. Limitations of vitamin D supplementation trials: Why observational studies will continue to help determine the role of vitamin D in health. J Steroid Biochem Mol Biol 2018;177:6–9. https://doi.org/10.1016/j.jsbmb.2017.06.006.
[19] Kuhn T. The structure of scientific revolutions. Chicago: University of Chicago Press; 1962.
[20] Holick MF. Vitamin D status: measurement, interpretation, and clinical application. Ann Epidemiol 2009;19:73–8. https://doi.org/10.1016/j.annepidem.2007.12.001

[21] Chesney RW, Rosen JF, Hamstra AJ, Smith C, Mahaffey K, DeLuca HF. Absence of seasonal variation in serum concentrations of 1,25-dihydroxyvitamin D despite a rise in 25-hydroxyvitamin D in summer. J Clin Endocrinol Metab 1981;53:139–42. https://doi.org/10.1210/jcem-53-1-139.
[22] Hine TJ, Roberts NB. Seasonal variation in serum 25-hydroxy vitamin D3 does not affect 1,25-dihydroxy vitamin D. Ann Clin Biochem 1994;31(Pt 1):31–4. https://doi.org/10.1177/000456329403100105.
[23] Biancuzzo RM, Clarke N, Reitz RE, Travison TG, Holick MF. Serum concentrations of 1,25-dihydroxyvitamin D2 and 1,25-dihydroxyvitamin D3 in response to vitamin D2 and vitamin D3 supplementation. J Clin Endocrinol Metab 2013;98:973–9. https://doi.org/10.1210/jc.2012-2114.
[24] Zittermann A, Ernst JB, Birschmann I, Dittrich M. Effect of vitamin D or activated vitamin D on circulating 1,25-dihydroxyvitamin D concentrations: a systematic review and metaanalysis of randomized controlled trials. Clin Chem 2015;61:1484–94. https://doi.org/10.1373/clinchem.2015.244913.
[25] Keppel Hesselink JM. Fundamentals of and critical issues in lipid autacoid medicine: a review. Pain Ther 2017;6:153–64. https://doi.org/10.1007/s40122-017-0075-4.
[26] Serhan CN, Levy BD. Resolvins in inflammation: emergence of the pro-resolving superfamily of mediators. J Clin Invest 2018. https://doi.org/10.1172/JCI97943.

[27] Hollis BW, Wagner CL. Clinical review: the role of the parent compound vitamin D with respect to metabolism and function: why clinical dose intervals can affect clinical outcomes. J Clin Endocrinol Metab 2013;98:4619–28. https://doi.org/10.1210/jc.2013-2653.
[28] Bouillon R, Garmyn M, Verstuyf A, Segaert S, Casteels K, Mathieu C. Paracrine role for calcitriol in the immune system and skin creates new therapeutic possibilities for vitamin D analogs. Eur J Endocrinol 1995;133:7–16.
[29] Ellfolk M, Norlin M, Gyllensten K, Wikvall K. Regulation of human vitamin D(3) 25-hydroxylases in dermal fibroblasts and prostate cancer LNCaP cells. Mol Pharmacol 2009;75:1392–9. https://doi.org/10.1124/mol.108.053660.
[30] Bonnet L, Hachemi MA, Karkeni E, Couturier C, Astier J, Defoort C, et al. Diet induced obesity modifies vitamin D metabolism and adipose tissue storage in mice. J Steroid Biochem Mol Biol 2019;185:39–46. https://doi.org/10.1016/j.jsbmb.2018.07.006.
[31] Landel V, Stephan D, Cui X, Eyles D, Feron F. Differential expression of vitamin Dassociated
enzymes and receptors in brain cell subtypes. J Steroid Biochem Mol Biol 2018;177:129–34. https://doi.org/10.1016/j.jsbmb.2017.09.008.
[32] Zehnder D, Bland R, Williams MC, McNinch RW, Howie AJ, Stewart PM, et al. Extrarenal expression of 25-hydroxyvitamin d(3)-1 alpha-hydroxylase. J Clin Endocrinol Metab 2001;86:888–94. https://doi.org/10.1210/jcem.86.2.7220.
[33] Hewison M. Vitamin D and immune function: autocrine, paracrine or endocrine? Scand J Clin Lab Invest Suppl 2012;243:92–102. https://doi.org/10.3109/00365513.2012.682862.
[34] Feldman D, Krishnan AV, Swami S, Giovannucci E, Feldman BJ. The role of vitamin D in reducing cancer risk and progression. Nat Rev Cancer 2014;14:342–57. https://doi.org/10.1038/nrc3691.
[35] El-Atifi M, Dreyfus M, Berger F, Wion D. Expression of CYP2R1 and VDR in human brain pericytes: the neurovascular vitamin D autocrine/paracrine model. NeuroReport 2015;26:245–8. https://doi.org/10.1097/WNR.0000000000000328.
[36] Dreyfus M, Wion D. Investigating the relationship between vitamin D and cancer requires dosing the bioavailable nonhydroxylated vitamin D storage in cancer tissues. Cancer 2015;121:3362–3. https://doi.org/10.1002/cncr.29451.
[37] Mawer EB, Backhouse J, Holman CA, Lumb GA, Stanbury SW. The distribution and storage of vitamin D and its metabolites in human tissues. Clin Sci 1972;43:413–31.
[38] Fu X, Dolnikowski GG, Patterson WB, Dawson-Hughes B, Zheng T, Morris MC, et al. Determination of vitamin D and Its metabolites in human brain using an ultrapressure LC-tandem mass spectra method. Curr Dev Nutr 2019;3:nzz074. https://doi.org/10.1093/cdn/nzz074.
[39] Safaiyan S, Kannaiyan N, Snaidero N, Brioschi S, Biber K, Yona S, et al. Age-related myelin degradation burdens the clearance function of microglia during aging. Nat Neurosci 2016;19:995–8. https://doi.org/10.1038/nn.4325.
[40] Neveu I, Naveilhan P, Menaa C, Wion D, Brachet P, Garabédian M. Synthesis of 1,25-dihydroxyvitamin D3 by rat brain macrophages in vitro. J Neurosci Res 1994;38:214–20. https://doi.org/10.1002/jnr.490380212.
[41] Yeung MSY, Zdunek S, Bergmann O, Bernard S, Salehpour M, Alkass K, et al. Dynamics of oligodendrocyte generation and myelination in the human brain. Cell

2014;159:766–74. https://doi.org/10.1016/j.cell.2014.10.011.
[42] Cui X, Gooch H, Petty A, McGrath JJ, Eyles D. Vitamin D and the brain: genomic and non-genomic actions. Mol Cell Endocrinol 2017;453:131–43. https://doi.org/10.1016/j.mce.2017.05.035.
[43] Garcion E, Wion-Barbot N, Montero-Menei CN, Berger F, Wion D. New clues about vitamin D functions in the nervous system. Trends Endocrinol Metab 2002;13:100–5.
[44] Lu M, Taylor BV, Körner H. Genomic effects of the vitamin D receptor: potentially the link between vitamin D, immune cells, and multiple sclerosis. Front Immunol 2018;9:477. https://doi.org/10.3389/fimmu.2018.00477.