



Antioxidants may increase the probability of developing allergic diseases and asthma

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Summary In addition to genetic predisposition, a lack of triggers for Th1 immune response like exposure to infections, endotoxins and dirt in childhood are supposed to be responsible for the higher incidence of allergic rhinitis and asthma (hygiene hypothesis). In vitro, beverages rich in antioxidants like green tea and wine were found to suppress formation of Th1-type cytokine interferon- γ . Due to the existing cross-regulatory interplay between Th1- and Th2-type immune response, these beverages may thus slow-down Th1-type immune response and thereby favour an over-production of Th2-type cytokines. Also food rich in antioxidants may increase the risk of atopic disease. Thus, not only a lack of triggers for Th1 type immune response, but also a nutrition rich in antioxidants suppressing interferon- γ would result in a persistence of Th2-type immune response and increase the susceptibility for allergic reactions and asthma. In addition to improved hygienic standards in the past decades, also social changes including the availability of functional food and food enriched in antioxidants may have increased the prevalence of atopic diseases in Western countries.

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Introduction

The incidence of allergic rhinitis and asthma is increasing. The possible reasons for this phenomenon are a topic of broad discussion. It seems that in addition to genetic predisposition, the strongest risk factors for development of these diseases may comprise several other causalities such as lifestyle

changes, atmospheric pollution, diesel fumes, maternal smoking, and greater exposure to dust mites [1,2]. The current understanding of asthma immunopathogenesis clearly relies on the predominance of Th2-type immune response suggesting over-production of Th2-type cytokines including interleukin-13 (IL-13) to be involved [3,4]. In this connection also the so-called "hygiene hypothesis" was established [5], which favours the view that a lack of triggers for Th1-type immune response like exposure to infections, endotoxins and dirt in childhood would result in a preponderance of Th2-type

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immune responses responsible for allergic disease [5,6].

Th1-type immune response and oxidative stress

Th1-type immune response is a most important part of adaptive immunity which is directed against host cells carrying non-self surface structures such as virus infected or malignant cells. Within Th1-type immune response, production of several cytotoxic compounds among them reactive oxygen species (ROS) such as hydrogen peroxide, superoxide anion or hydroxyl radical, are released by macrophages as central part of their antimicrobial and cytotoxic armature (Fig. 1), the most potent trigger being interferon- γ (IFN- γ) [7]. During sustained periods of Th1-type immune activation, e.g., in chronic infections, the continuing production of these oxidizing species may destroy antioxidant pools and oxidative stress is developing as a consequence. An oxidizing milieu is also a relevant trigger of redox-sensitive signal transduction pathways in cells including the induction of pro-inflammatory cytokines like TNF- α via the nuclear transcription factor NF- κ B, a central and redox-sensitive mediator of

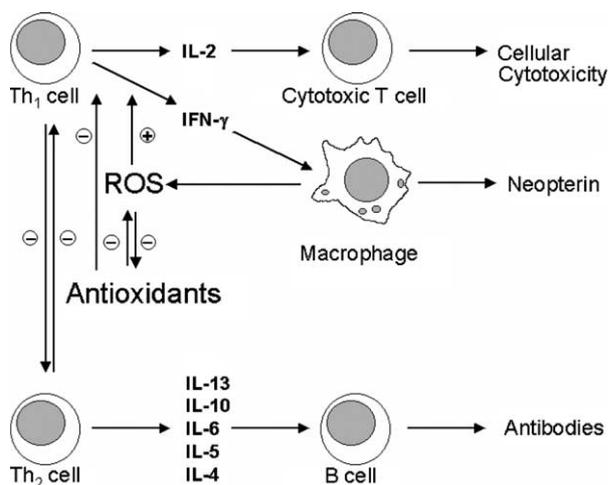


Figure 1 Th1-type immune response, characterized by cytokines like interleukin-2 (IL-2) and interferon- γ (IFN- γ) is accompanied by the formation of neopterin and reactive oxygen species (ROS) by macrophages and results in specific cellular cytotoxicity. Th2-type immune response, characterized by cytokines like IL-4, -5, -6, -10, -13 results in the formation of specific antibodies (=immunoglobulins). There exists a cross-regulatory interplay between Th1-type and Th2-type immune response, down-regulating each other when activated. Antioxidants may weaken the effect of ROS and inhibit Th1-type cytokine production.

inflammation [8,9]. Also the production of IFN- γ is inducible by oxidizing agents such as hydrogen peroxide [10]. Thus, the production of ROS not only is part of the killing strategy of effector cells within the Th1-type immune response, it is also involved to further amplify the release of pro-inflammatory cytokines (Fig. 1).

In vitro studies show a cross-regulatory influence between Th1- and Th2-type immune responses, down-regulating each other when activated, e.g., typical Th2-type cytokines like IL-4 and IL-10 inhibit the production of Th1-derived cytokines such as IFN- γ and vice versa [11]. Because of this cross-regulatory interplay, the susceptibility for allergic diseases may not only increase when Th2-type cytokines are over-produced but also when Th1-type cytokines such as IFN- γ are suppressed (Fig. 1).

Pro-inflammatory cytokine IFN- γ is probably the most important mediator of anti-microbial and anti-tumoral defence, e.g., it induces high-output of cytotoxic ROS in macrophages and other cells [7]. IFN- γ is also primary for the expression of enzymes indoleamine (2,3)-dioxygenase (IDO) and GTP-cyclohydrolase I [12]. Activation of the latter is reflected by increased neopterin concentrations in patients during diseases which are associated with Th1-type immune activation [13], activation of IDO can be demonstrated in the same patients by the increased conversion of tryptophan to kynurenine derivatives and increased kynurenine to tryptophan ratio (kyn/trp) [14]. In vivo investigations also confirm the cross-regulatory interplay between Th1-type and Th2-type immune response, e.g., an inverse relationship exists between immunoglobulin E concentration, typically for Th2 type immune response, and neopterin, which is released in increased amounts from macrophages on stimulation with IFN- γ , and is thus typical for Th1-type immune response [15,16]. In addition it could be shown that increased pulmonary IDO activity, which may be due to Th1-type immune activation or innate immune activation, inhibits Th2-driven experimental asthma in a murine ovalbumin sensitization model [17].

Antioxidants suppress Th1-type cytokine IFN- γ

Measurement of neopterin production and of tryptophan degradation can also be applied to monitor Th1-type immune response in vitro, e.g., in peripheral blood mononuclear cells (PBMC) stimulated by mitogens phytohaemagglutinin and concanavalin A. Thereby it is possible to determine effects of stim-

ulatory or suppressive compounds, e.g., Th2-type cytokines IL-4 and IL-10 significantly reduce neopterin production and kyn/tryptophan in supernatants of PBMC [12]. Accordingly, also histamine, an important mediator of Th2 cell-driven allergic reaction released from mast cells, significantly suppresses neopterin formation [18]. Using this in vitro assay, nutrients and alcoholic and alcohol-free beverages rich in antioxidants such as green tea and wine and also plant extracts were found to interfere with pathways involved in Th1-type immune response [19–22]. Antioxidant content of these beverages appeared to be important for their down-regulatory capacity of Th1-type immune response. Antioxidants achieve a similar effect as found for histamine, however, histamine content of beverages is too low and cannot be fully responsible for these findings.

Also aspirin (acetylsalicylic acid) which is widely used in the treatment of inflammatory disorders mainly because of its inhibitory effects on cyclooxygenase I and II was found to inhibit neopterin production and tryptophan degradation in stimulated PBMC [23]. Since aspirin also possesses antioxidant properties [24–26], antioxidative and radical-scavenging activity could be important for its down-modulatory effect on IFN- γ production in stimulated PBMC. This assumption may be further encouraged by the observation that aspirin may induce asthma in some individuals [27]. In line with these findings, astaxanthin, a carotenoid without provitamin A activity, was found to suppress IFN- γ production in a Th1 cell clone and, in parallel, increased the number of antibody-secreting cells [28]. Also from in vivo investigations the influence of oxidants and antioxidative capacity on immune activation seems to be probable. On the one hand, a positive correlation of neopterin formation and advanced oxidation protein products in uremic patients was found [29], on the other hand, an inverse correlation between neopterin production and serum vitamin E concentrations in nonagenarians was observed [30]. In conclusion, there is ample indication that antioxidants may suppress Th1-type immune activation by reducing oxidative capacity, which in turn enhances immunoglobulin production by Th2-type cells.

“Healthy food” rich in antioxidants increases the susceptibility for allergy and asthma

Antioxidants inhibiting Th1-type immune activation, may promote Th2-type immunity and thereby

increase the susceptibility for allergic diseases and asthma when an allergen is incorporated. In contrast, ROS are not only produced in case of Th1-type immune response, they may also promote cellular cytotoxicity by induction of Th1-type cytokines like IFN- γ , thereby down-regulating Th2-type immune response (Fig. 1). In other words, a disturbance of the equilibrium between pro- and antioxidants in the organism could determine – in addition to possible other factors – which type of specific immune reaction mediated by Th1- or Th2-type cells would be favoured (Fig. 2). “**Antioxidative stress**” may result from too much intake of food rich in antioxidants, and it could increase the susceptibility for allergic reactions and asthma by down-regulating Th1-type immune response and thereby promoting Th2-type cells and immunoglobulin production. A higher oxidative capacity might favour Th1-type immune response and lower the susceptibility for allergic reactions by inhibiting immunoglobulin production. Consequently, not only to little challenge by infectious agents but also too much of “healthy food” rich in antioxidants would increase the susceptibility for allergy and asthma.

From these data it is tempting to extend the “hygiene hypothesis”: not only a lack of triggers for Th1-type immune response like exposure to infections, endotoxins and dirt in childhood would result in a persistence of Th2-type immune

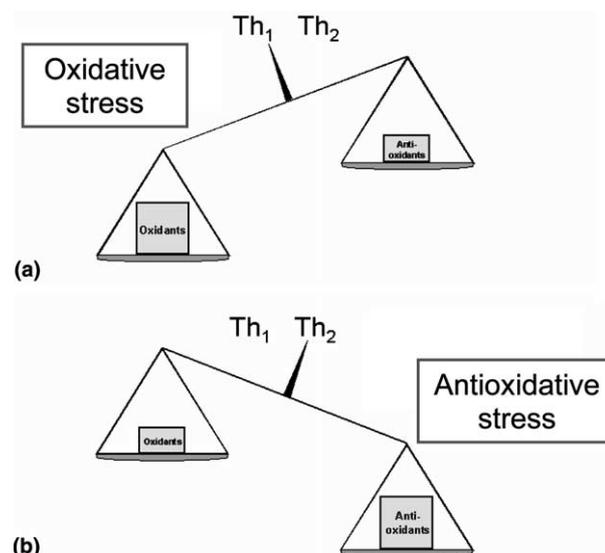


Figure 2 Influence of the balance of oxidants and antioxidants on the type of immune activation. In case of excessive oxidants (oxidative stress) Th1-type immune response dominates (a). Excess of antioxidants (antioxidative stress) may favour Th2-type immune response (b).

response, but also a nutrition rich in antioxidants might increase the susceptibility for allergic reactions and asthma. This hypothesis may specifically relate to the increased frequency of allergic reactions against citric fruit allergens, which are also assumed to be responsible for specific allergic reactions in certain individuals [31,32].

Contradictions and perspectives

Studies on the prevalence of asthma showed that a frequent consumption of fruit and raw vegetables would reduce risk of asthmatic symptoms like, e.g., wheezing [33]. Also naturally occurring polyphenolic antioxidants were described earlier to modulate IgE-mediated mast cell activation [34], and ROS were suggested to enhance allergic inflammation [35]. At first glance, such observations might contradict our hypothesis, and obviously the strict counterbalance between Th1- and Th2-type immune response certainly is too simplistic, e.g., allergen-specific Th1 cells were found to fail to counterbalance Th2 cell-induced airway hyper-reactivity but can cause severe airway inflammation [36]. On the other hand, it should be considered that the composition of fruits or vegetables is rather complex, the effect of many of its components on human organism is not jet clear in detail and that the ultimate biologic activity of given food depends on large number of variables, including, e.g., food processing, preparation method, interactions between compounds in the food [37]. Finally, *in vitro* antioxidants sometimes may produce artificial results, since they may reduce air oxygen to produce reactive species such as superoxide anion. In addition, different culture media may have different effect, e.g. the ability of ascorbate to interact with different cell culture media to produce hydrogen peroxide could lead to conflicting results [38].

Prospective studies comparing levels of antioxidants, protein oxidation products in blood should be analyzed in patients with allergic diseases and asthma. Correlations thereby found might confirm the hypothesis but are unable to prove it. However, there are several other possibilities, allergic patients and healthy controls should undergo controlled supplementation with antioxidants and the effect on Th1 cell and Th2 cell activation should be measured by, e.g., neopterin concentrations and antibody titers, respectively. Also correlations with the activity of allergic diseases should be established in the same study design. Animal models might allow to test whether antioxidants are

able to direct immune response to, e.g., airway allergy. However, only detailed examinations of the mechanisms by which antioxidants influence immune response will allow to confirm or weaken this hypothesis. It should be considered that typical antioxidants may influence signal transduction pathways not only by their reducing effect on oxidants but also by other so far unknown mechanisms.

In conclusion, additionally to improved hygienic standards in the past decades, also social changes including the availability of functional food and food enriched in antioxidants may have increased the prevalence of atopic diseases in Western countries. The "hygiene hypothesis" might be biased by other lifestyle-related changes, because improved lifestyle not only has contributed to decrease infectious challenges, it also improved food standards.

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