



Immunological models in high dilution research following M Bastide

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In 1994, Madeleine Bastide described experimental models in immunology that were used during the 1980s to investigate high dilution effects on several biological systems. She classified the available papers in four categories: High dilutions of antigens; High dilutions of thymus, bursa and other hormones; High dilutions of cytokines; Immunopharmacological activity of silica. The studies about high dilutions of antigens were not continued after this period, but gave rise to a long process of a series of *in vitro* models on antigens and histamine dilutions, that led to the demonstration of the biological modulation effects of these preparations on basophil degranulation. During this process, a multi-centre study was performed, with a high degree of reproducibility among different independent laboratories.

The studies about high diluted cytokines, thymulin and other hormones opened a new line of scientific investigation, about the regulatory properties of endogenous substances prepared according to homeopathic methods. The most frequently studied substance, thymulin, when administered to mice at 5cH potency, is able to improve the activity of phagocytes in different experimental situations, such as viral, bacterial and parasitic infections. The immunopharmacological activity of silica was demonstrated, at that time, as an *in vivo* illustration of the homeopathic 'similia principle'. More recently, studies on silica have assumed another focus: the putative role of silica as active contaminant present in high dilutions. This paper presents a follow-up summary on these items, considering the evolution of discoveries from 1994 to 2014. *Homeopathy* (2015) 104, 263–268.

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Introduction

In her seminal 1994 review,¹ the late lamented Prof. Madeleine Bastide (Figure 1) showed that immunological models are useful for demonstrating the activity of homeopathic remedies or effects of high dilutions of hormones or mediators. The whole living organism is able to respond to homeopathic remedies as well as to antigens. Of course, the immunological response is not mechanistically comparable to the action of a homeopathic remedy, but the immune system is naturally stimulated by extremely low doses of antigens (as low as 0.1 ng [10^{-10} g]²). In an *in vitro* model, human monocytes have been shown to present optimal phagocytosis using 10^{-14} M postin, and

such activity was still evident at 10^{-18} M.³ Evaluations of the effects of high dilutions or very low doses in immunological models are still an interesting methodological possibility and open a new field of application using homeopathic preparations in both the therapeutic and agricultural fields.

Methods

The main sources of information for this review were personal contacts with members of the homeopathic research community, mainly during the annual symposium of the *Groupe International de Recherche sur l'Infinitésimal* (GIRI; <http://www.giriweb.com>; accessed May 21, 2015), and the MEDLINE and HOMBREX databases. Experimental studies that used animal models were extracted from the publications and grouped into models. No clinical or human-based research data were considered in this review.

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Figure 1 Prof. Madeleine Bastide (1935–2007). For bibliography, see Ref. 52.

Results

High dilutions of antigens

The original discussion concentrated on the work of Weisman and colleagues, 1992,⁴ in which high dilutions (10^{-15} – 10^{-33} M) of KLH protein (keyhole-limpet-hemocyanin) that are prepared in centesimal potencies, including those above Avogadro's number, were able to signal the immune system similarly to the antigen at measurable concentrations and induce the production of polyclonal immunoglobulin M (IgM). After a new immunization, a specific potency-dependent increase in monoclonal anti-KLH immunoglobulin G (IgG; an immune memory antibody) could be seen.

After this period, new articles on immune stimulation with highly diluted substances focused on allergy research. In 2000, Aabel and colleagues^{5,6} discussed the curative and prophylactic efficacy of *Betula* 30C in clinical trials. No prophylactic effect was found, but a curative effect was observed. The benefits of homeopathy's curative approach for treating allergies were reinforced by other authors a few years later.^{7–9}

Most studies that utilized this model discussed the modulation of *in vitro* human basophil degranulation by highly diluted antigens, such as *Apis mellifica*¹⁰ and histamine.¹¹ After 20 years of discussion, Sainte-Laudy and Belon published their final conclusion about the effectiveness of 15–17cH potencies of histaminum as a reproducible biological effect, which was confirmed by a large multicenter study¹¹ and most independent authors.^{12,13} The effect of highly diluted/succussed histamine presented high specificity. The same potencies of related nonspecific substances, such as histidine, had no effect and the histamine action was blocked by the H₂ receptor inhibitor cimetidine. The authors also discussed the putative role of histamine H₂ receptors in the mechanism of action.¹⁴ The reported difficulties of reproducibility in different laboratories suggest that this model may serve

to identify possible technical factors in the manifestation of the regulating power of high dilutions of histamine.

High dilutions of thymulin, bursin, and other hormones

Bastide, in 1994¹ discussed her own work on high dilutions of thymulin and bursin. Her first experiment was mentioned, in which C57/BL6 mice with P815 mastocytoma were treated with different centesimal potencies of thymulin and exhibited a clear immune depressive effect, with decrease in the specific anti-tumor activity of T cells.^{15,16} Another series of experiments that were performed by^{17,18} were also mentioned, showing that thymulin 9C modulated the T-cell dependent humoral antibody response and respective corticosterone serum levels in mice, with an emphasis on seasonal variations in these effects.

Another hormone that was studied by Bastide was bursin. In a sophisticated experimental model that was developed by Guellati, in 1990¹⁹ and Youbicier-Simo and colleagues, in 1993,²⁰ chicken embryos were surgically deprived of *Fabricius bursa* on the day 4 of incubation, and their specific B cell response recovered after *in ovo* treatment with different dilutions of bursin up to 10^{-40} M. The IgM and IgG titrations were two-times those seen in controls. This study demonstrated how patterns of biological functions can be programmed by the transfer of information from high dilutions to living systems and how developing tissues are sensitive to this.

New studies on homeopathic preparations of hormones were continued into the next decade. In 2001, Bonamin and colleagues²¹ performed a study that demonstrated the capacity of dexamethasone 7 and 15cH (10^{-17} and 10^{-33} M, respectively) to negate the anti-inflammatory effect of pharmacological concentrations of the same hormone using a classic experimental model of inflammation in mice. Some years later, effects on inflammation were also shown in adult rats that were born from mothers that were treated with dexamethasone 15cH during pregnancy. This was the first demonstration in rats of the mother-newborn transmission of homeopathic information and its ability to imprint functional 'patterns' on the physiology of offspring during the perinatal period.²²

Considering the effects of homeopathic thymulin, new experiments were subsequently performed after Bastide's publications. Sato and colleagues, in 2012,²³ showed the usefulness of thymulin 5cH diluted in drinking water (20 ml of thymulin 5cH in 50,000 ml of water) to improve weight gain, viability (from 75% to 97.5%), the cellular immune response against viruses (22%), and quality of life of broiler chickens (reflected by the productivity index; 22%) compared with controls, suggesting the use of homeopathic thymulin as a useful zootechnical tool, as a promoter of animal welfare.

In addition to the known improvement of the T-cell response that was described in mice and chickens, a more recent study reported the effects of thymulin 5cH on the recruitment of B1 cells (CD19+, CD11b+, and CD23– stem cells) from the peritoneum to local

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