A perspective of nanotechnology in hypersensitivity reactions including drug allergy

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Purpose of review
We provide an overview of the application of the concepts of nanoscience and nanotechnology as a novel scientific approach to the area of nanomedicine related to the domain of the immune system. Particular emphasis will be paid to studies on drug allergy reactions.
Recent findings
Several well defined chemical structures arranged in the dimension of the nanoscale are currently being studied for biomedical purposes. By interacting with the immune system, some of these show promising applications as vaccines, diagnostic tools and activators/effectors of the immune response. Even a brief listing of some key applications of nanostructured materials shows how broad and intense this area of nanomedicine is.
Summary
As a result of the development of nanoscience and nanotechnology applied to medicine, new approaches can be envisioned for problems related to the modulation of the immune response, as well as in immunodiagnosis, and to design new tools to solve related medical challenges. Nanoparticles offer unique advantages with which to exploit new properties and for materials to play a major role in new diagnostic techniques and therapies. Fullerene-C60 and multivalent functionalized gold nanoparticles of various sizes have led to new tools and opened up new ways to study and interact with the immune system. Some of the most versatile nanostructures are dendrimers. In their interaction with the immune system they can naturally occurring macromolecules, taking advantage of the fact that dendrimers can be synthesized into nanosized structures. Their multivalence can be successfully exploited in vaccines and diagnostic tests for allergic reactions.

Keywords
dendrimers, drug allergy, nanomedicine, nanoparticle, nanotechnology

Introduction

Nanoscience and nanotechnology have been described as the science involved in the study and design of ‘novel materials whose size of elemental structure has been engineered at the nanometre scale’. Materials in the nanometre size range commonly exhibit fundamentally new behaviour.

Nanotechnology is an area that has seen a surge in research activity over recent years. It is considered to be one of the emerging fields in science, with great potential in a wide range of applications. Structures on the nanoscale allow materials to exhibit different proper- ties, owing to their size. The development of these tools and of new materials has the potential to provide novel approaches, improving current therapeutic and diagnostic tools and eventually addressing a range of unmet medical needs.

Nanoscience and nanotechnology

The concept of nanoscience concerns a collection of different disciplines and techniques involved in the study and manipulation of matter at the nanometric scale. At this scale, in which the frontiers between physics, chem- istry and biology overlap, an interdisciplinary and con- vergent approach of this new branch of science is favoured. The nanometre scale ranges from the atomic level, at around 0.1 nm (1 A˚), up to around 100 nm (Fig. 1).

Nanotechnology is a broad term covering a wide range of methods, tools and possible applications. A variety of definitions have been given in the literature, each generated for different purposes. Nanotechnology refers to the top-down and bottom-up manipulation of matter down to the atomic level, ranging from atoms, molecules and biomolecules, to particles and structured supramo- lecular assembling, to reach functional building blocks. Nanotechnology in health and medical systems is part of what has been termed 'nanobiotechnology'. It combines biological and inorganic matter at atomic and molecular level, resulting in the possible creation of new matter with new functional properties. The main areas of application lie within biology, medicine and medical technology.

Nanomedicine

Among the various areas in science, the rapid development of nanoscience and nanotechnology is expected to have a remarkable impact on medicine. Nanomedicine is a direct result of the application of nanotechnology to medicine in order to make a medical diagnosis or treat or prevent diseases. It exploits the improved and often novel proper- ties of materials at the nanometre scale. Nanomedicine is emerging as a new clinical field that applies nanotechnology to solve a wide range of current medical problems. New nanotechnology-based methods and materials with innovative nanoscale properties are being investigated and successfully developed in major areas of research, such as targeted drug delivery, medical nanomaterials, diagnosis, regenerative medicine and biomaterial implants, which have already yielded favourable responses.

Nanoparticles

Several varieties of nanoparticles exist such as different polymeric and metal nanoparticles, liposomes, micelles, quantum dots, dendrimers, microcapsules, cells, cell ghosts, lipoproteins and many different nanoassemblies. All these nanoparticles are able to play a major role in different areas of medical diagnosis and therapy. Here we present relevant contributions to topics related with the immune system, from diagnosis to vaccines. Nanoparti- cles, frequently composed of multiple components with varying compositions, sizes and surface properties, may be engineered to either avoid immune system recog- nition or specifically inhibit or enhance the immune responses. Manipulation of nanoparticle size and charge has proven useful to increase particle delivery to immune cells for applications such as vaccine delivery.[1].

Fullerene derivatives

Fullerenes, one form of nanomaterial, are novel carbon allotropes forming soccer ball-shaped carbon cages (C60) that can be functionalized and derivatized with a wide array of molecules. Ryan et al. [2] reported fullerenes as a negative regulator of allergy mediator release. Human mast cell and peripheral blood basophils exhibited a sig- nificant inhibition of immunoglobulin E (IgE)-dependent mediator release when preincubated with C60 fullerenes. This involved profound reductions in the activation of signalling molecules involved in mediator release and oxidative stress, and it significantly inhibited IgE-induced elevation in cytoplasmic reactive oxygen species. Further- more, fullerenes prevented the in-vivo release of hista- mine and a drop in core body temperature in vivo using a mast cell-dependent model of anaphylaxis. Future appli- cations can be guided towards the study of fullerenes modified with functional groups to specifically target their uptake to mast cell and peripheral blood basophils [3] (Fig. 2a).

Metallic nanoparticles

A well designed demonstration of the relevance of the size-to-multivalence relationship has been shown by the possibility of obtaining engineered nanoparticles of well defined sizes and surface ligand densities that can selec- tively control the degree of receptor cross-linking, which, in turn, alters downstream signalling and subsequent cell responses.

Huang et al. [4] showed that it is possible to produce altered signal transduction by synthetic means, developing chemi- cally defined multivalent ligands of effectors. Specifically, the average spacing between two binding sites within an antibody and the average distance between receptors on the cell membrane could either promote or inhibit cell activation. These authors directly addressed these chal- lenges by demonstrating how gold nanoparticles of pre- cisely controlled mean diameters can be easily synthesized and surface-modified at an equally well controlled ligand density or spacing. These authors found that both nano- particle size and surface ligand density play key regulatory roles in the process of membrane antibody-receptor bind- ing and cross-linking, which, in turn, leads to degranulation and subsequent release of chemical mediators. These results demonstrate that nanoparticles serve not only as simple platforms for multivalent binding but also as mediators for key biological functions (Fig. 2b).

Dendrimers
Dendrimers may well represent the nanosized structures that have opened up more expectations for biomedical applications. They consist of very well defined, mono- disperse, stable molecular level nanostructures and are studied according to their ‘dendritic state’ architecture. Dendrimers may be visualized as consisting of three critical architectural domains: (a) the multivalent surface, containing a large number of potentially reactive and/or passive sites, (b) the interior shells surrounding the core and (c) the core to which the branches are attached [5]. The three traditional macromolecular architectural classes (linear, cross-linked and branched) are widely recognized to generate rather polydisperse products of different molecular weights. In contrast, the synthesis of dendrimers offers the opportunity to generate monodis- perse, structure-controlled macromolecular architectures similar to those observed in biological systems [6] (Fig. 3).

Dendrimer vaccines
Dendrimers have optimal characteristics to fill the need for efficient immunostimulating compounds (adjuvants)
that can increase the efficiency of vaccines, as they can provide molecularly defined multivalent scaffolds to produce highly defined conjugates with small molecule immunostimulators and/or antigens, enabling knowledge- based vaccine design in substitution of the traditional empirically based approaches for vaccine development and production [7●●].

Dendrimers have properties of multivalency, size and structural definition that allow them to be used as build- ing blocks, scaffolds or carriers for creating efficient vaccine components that will induce a desired and pre- determined type of immunity in the vaccinated host. Traditionally, carrier molecules have been nonself (foreign) proteins used to bind small, nonimmunogenic antigens to make them immunogenic. Using these meth- odologies it is not possible to predict optimal carrier, conjugation chemistry and immunogenic-carrier coupling ratios for desired immune responses. In addition, the conjugates cannot be easily analysed, they have unknown coupling ratios and unknown coupling orientation of the antigen and they may contain impurities consisting of polymers or oligomers of the antigens and the carrier protein. Furthermore, naturally derived carrier proteins are not fully chemically characterized or standardizable and may be expensive.
The preparation of highly defined, reproducible immu- nogens for human vaccine use requires other more desir- able types of carrier. With this in mind, dendrimers have emerged as useful tools as they can act as multivalent and well defined carriers for antigenic substances by coupling of antigen molecules to the surface functional groups of the dendrimer.

Dendrimer diagnosis

At the molecular level, dendrimers are potential diag- nostic agents, thanks to the ease with which they can incorporate multiple functionalities leading to high activity through multiple interactions that result in increased sensitivity. Dendrimers containing multiple identical ligands are very attractive, as these structures can exhibit amplified substrate binding, for either stat- istical or cooperativity effects.

Dendrimeric antigens: hapten–dendrimer conjugates

A protein is necessary for the processing and production of IgE antibodies to b-lactams in order to become allergic [8]. Once subjects become allergic, a protein or any type of carrier that produce multivalent hapten-carrier con- jugates can be used to be recognised by IgE antibodies. Recently [9], the preparation and properties of polyami- doamine (PAMAM) dendrimers functionalized with peripheral benzylpenicilloyl (BPO) units have been reported. The open form of the b-lactamic antibiotics, the penicilloyl group, is the major determinant respon- sible for the allergic response to these drugs [10]. The synthesis of these BPO-containing dendrimers has been achieved by a convenient procedure involving quantitat- ive functionalization of the terminal amino groups of the three PAMAM generations used [9]. The general aim of this study was to design and synthesize a sequence of precisely defined molecular structures to achieve hap- ten–carrier conjugates of increasing BPO density at the periphery of preformed PAMAM dendritic cores and to exploit their exo-receptor properties in IgE detection.

The initial results with sera from patients with different RAST levels were positive and thus suggested that inhibition occurs, so recognition exists. These prelimi- nary RAST inhibition studies confirmed the usefulness of penicilloylated PAMAM dendrimers as conjugates for the recognition of IgE antibodies from patients allergic to penicillin. It can therefore be concluded that the hapten– carrier (dendrimer) conjugates studied mimic recognition with natural hapten–carrier (protein) conjugates.

Dendrimers in RAST for drug allergy diagnosis

The structural precision of dendrimers has prompted a number of studies aimed at developing new biochemical applications [11]. However, their potential for emulating the carrier protein in conjugates for IgE recognition has hardly been exploited. In the belief that dendrimers, based on their shape and physical properties [12], can be viewed as protein mimetics and have a potential as protein-like materials for biotechnological applications, penicilloylated dendrimers from generations 0– 6 were prepared on cellulose surfaces as platforms for detecting IgE antibodies from penicillin-allergic patients [13]. Recent progress in the production of a series of cellulose solid phases chemically functionalized with densely peni- cilloylated PAMAM dendrimers of increasing binding epitope density and size scaling has been reported. The use of this panel of dendrimers has also been evaluated for the recognition and quantification of IgE antibodies directed to benzylpenicillin, as has the effect of changes in ligand valency related to changes in epitope presentation (viz. residue density and architecture). The chemical methodology used in this study allows standar- dized materials with a definite total amount of attached dendrimers to be reproducibly prepared. The new material has been successfully used to detect IgE in patients diagnosed with an immediate allergic reaction to benzylpenicillin (Fig. 4). A more precise and realistic approximation will require considerations on the possible structure of the antigenic determinant formed in vivo, and although some fine-tuning will be required to establish an optimum diagnostic method, the RAST results obtained in this study suggest that dendrimerized cellulose discs meet all the requirements for the development of clini- cally useful diagnostic tests.

Conclusion

The possibility of manipulating matter at the nanoscale permits understanding of the relationship between structure at the nanometer dimension and new properties. This enables the preparation of new structures and assemblies whose intrinsic properties depend directly on their size [14●●]. The emergence of the recently recognized field of nanomedicine has led to useful applications in the area of immunology, searching for new and tuneable properties. Eligible devices that have already been used in this field include metallic nanoparticles, the allotropic form of car- bon-like fullerenes and, more importantly, dendrimers, which present the required properties for this purpose. Their unique structure, which provides them with stability, a spherical shape, controlled size, hollow struc- ture and the intrinsic property of multivalency, has led to dendrimers often being referred to as synthetic com- pounds able to mimic proteins in their interaction with the immune system. This success suggests they can be used for the in-vitro diagnosis of allergy instead of having to use invasive in-vivo tests, which are usually considered as the standard in the diagnosis of hypersensitivity reactions.

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References and recommended reading

Papers of particular interest, published within the annual period of review, have been highlighted as:
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Additional references related to this topic can also be found in the Current World Literature section in this issue (p. 402).

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