Propojení výuky oborů Molekulární a buněčné biologie a Ochrany a tvorby životního prostředí OPVK (CZ.1.07/2.2.00/28.0032)

Distinct types of diffuse large B-cell lymphoma identified by gene expression profiling

Martina Onderková

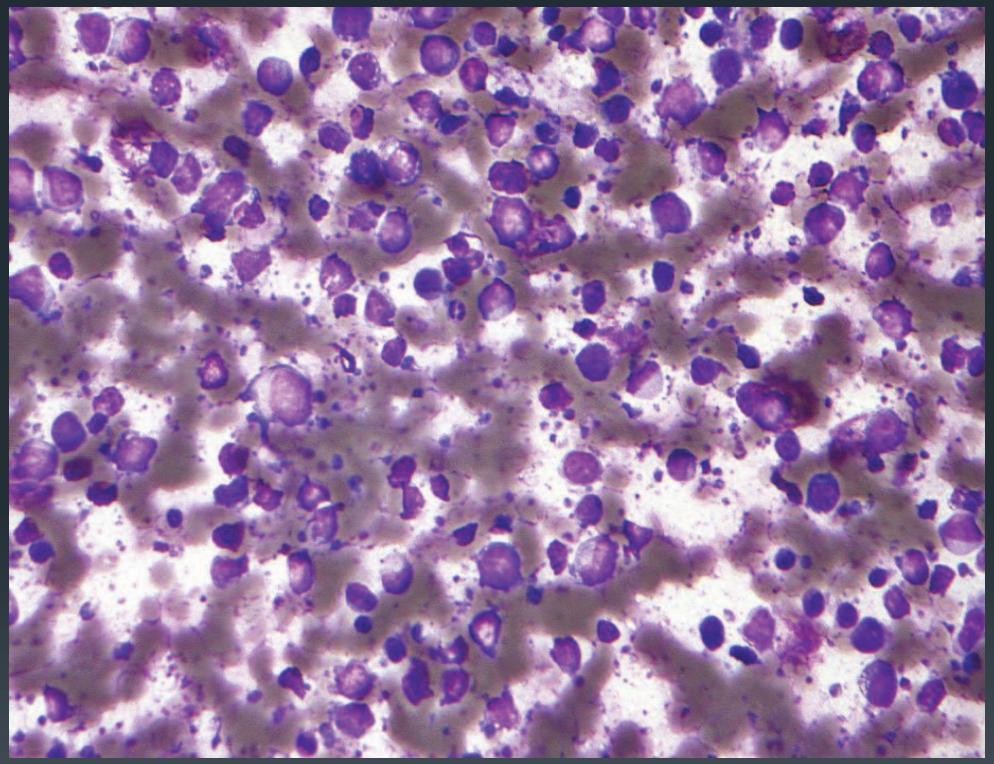
NATURE
VOL 403
3 february 2000

Ash A. Alizadeh

esf evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

What is it DLBCL?

- Cancer of B-cell
- Type of non-Hodgkin lymphoma among adults
- This cancer accurs primarily in older individuals
- aggressive tumour which can arise in virtually any part of the body



esf evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

Why examine this type of cancer?

- Clinically heteregeneous: 40 % of patients respond well to current therapy and have prolonged survival, whereas the remainder succumb to the disease.
- They proposed that this variability in natural history reflects unrecognized molecular heterogeneity in the tumours.
- Some patients receiving the same diagnosis can have markedly different clinical courses and treatment responses.

An important component of the biology of a malignant cell

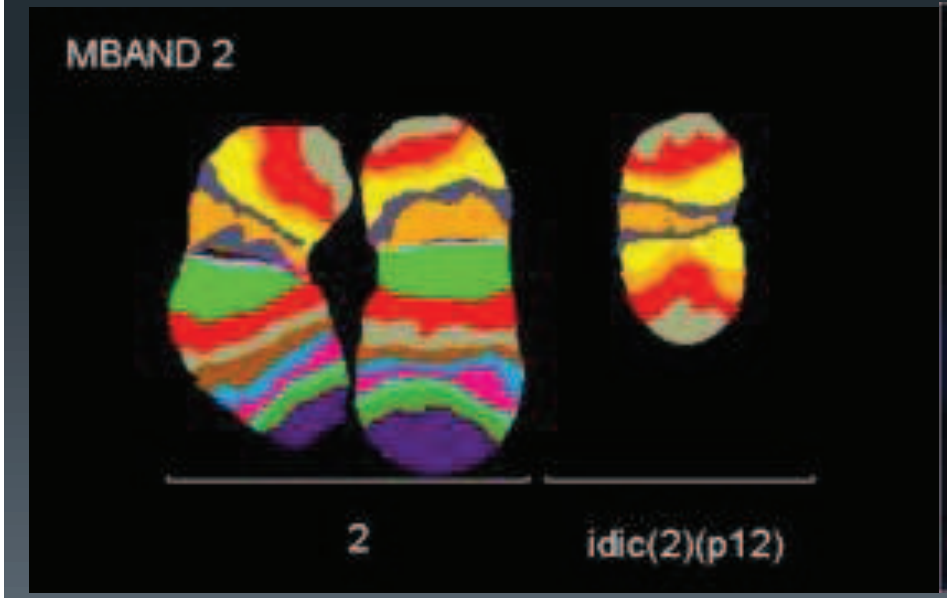
- is inherited from its non-transformed cellular progenitor
- Each of the currently recognized categories of B-cell malignancy has been tentatively traced to a particular stage of B-cell differentiation, although the extent to which these malignancies maintain the molecular and physiological properties of normal B-cell subsets is not clear.
- The rearranged immunoglobulin genes in DLBCL and most other non-Hodgkin's lymphomas bear mutations that are characteristic of somatic hypermutation, in antibody-diversification mechanism that normally occurs only within the germinal centre of secondary lymphoid organs.
- This evidence suggests that DLBCL arises either from germinal centre B cells or from B cells at a later stage of differentiation.

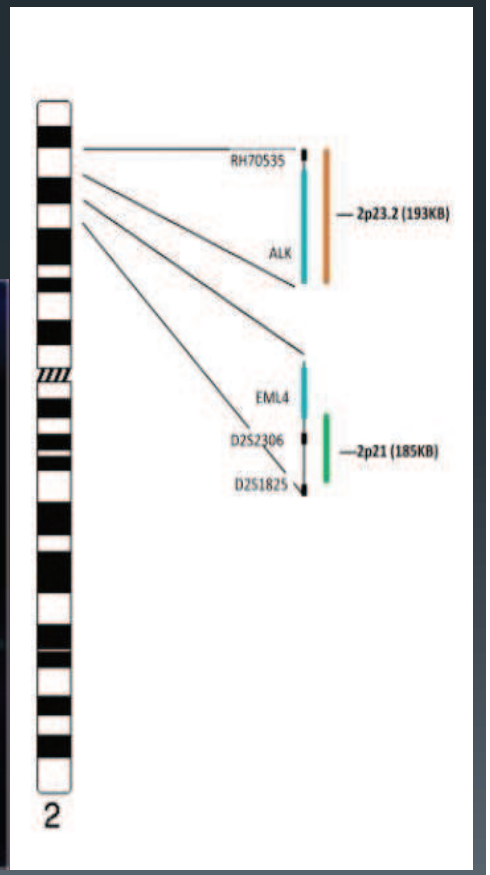
Metods

- They used DNA microarrays and they conducted a systematic charakterization of gene expression in B-cell malignancies.
- -> to questions in normal and malignant lymphocyte biology, designed a specialized microarray - **the `Lymphochip** - by selecting genes that are preferentially expressed in lymphoid cells and genes with known or suspected roles in processes important in immunology or cancer
- This metod show that there is diversity in gene expression among the tumours of DLBCL patients, apparently reflecting the variation in tumour proliferation rate, host response and differentiation state of the tumour.

- Samples from sick patients ←→ model samples = level of gene expression

- They also used fluorescent probes:





The result

- identified two molecularly distinct forms of DLBCL which had gene expression patterns indicative of different stages of B-cell differentiation:
- germinal centre B cells (**GL B-like DLBCL**)
- normally induced during *in vitro* activation of peripheral blood B cells (**activated B-like DLBCL**)

- Patients with germinal centre B-like DLBCL had a significantly better overall survival than activated B-like DLBCL.

Conclusions

- This study shows that a genomic view of gene expression in cancer can bring clarity to previously muddy diagnostic categories.
- The precision of morphological diagnosis, even when supplemented with immunohistochemistry for a few markers, was insufficient in the case of DLBCL to identify believable diagnostic subgroups. A number of individual markers have been used to define subsets of DLBCL, but these studies do not provide the present overview that strongly implies that this single diagnostic category of lymphoma harbours at least two distinct diseases.
- Indeed, the new methods of gene expression profiling call for a revised definition of what is deemed a `disease'.

esf evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

Contribution

- The molecular classification of tumours on the basis of gene expression can thus identify previously undetected and clinically significant subtypes of cancer and thus affect treatmen.

- connections with other disciplines (immunohistochemistry..)

- **Why is this topic still actual?**

- it´s very expensive and labor – intensive
- new studies

esf evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

Thank you for your attention!

Propojení výuky oborů Molekulární a buněčné biologie a Ochrany a tvorby životního prostředí OPVK (CZ.1.07/2.2.00/28.0032)

Balancing act

Monika Opatíková

Nature, vol 510, 19 June 2014

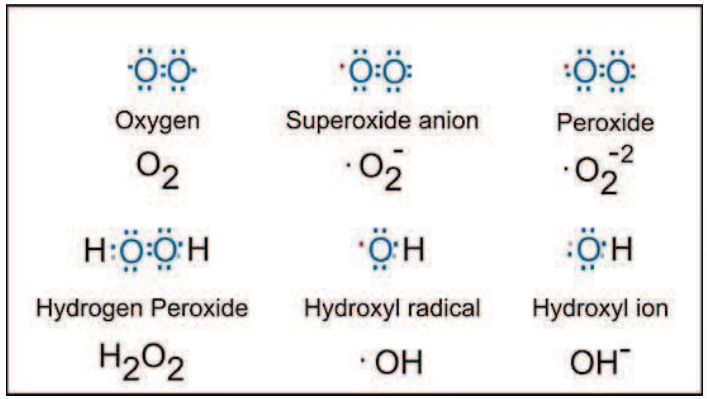
Alban Ordureau & J. Wade Harper

esf evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

Mitochondrion and its destruction

- enzyme parkin: disposal of damaged mitochondria
- opposes enzyme: balance violation

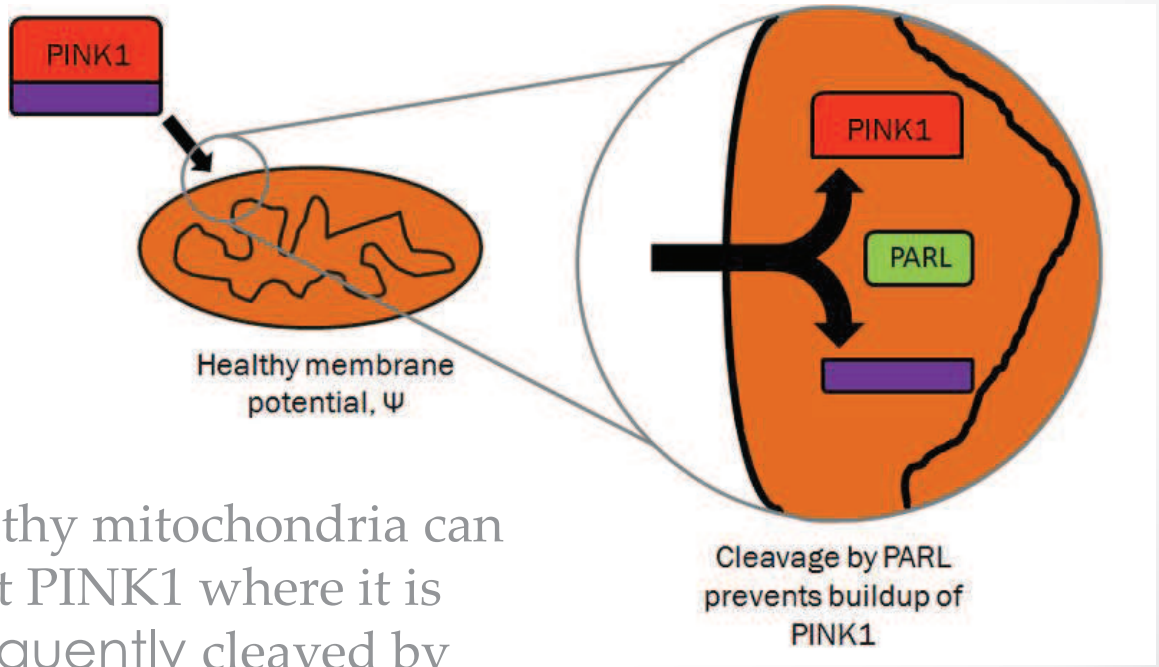
- mitochondria provide energy, but defects in this organelles can lead to the production of reactive oxygen species that disrupt crucial cellular function
- mitophagy: process that eliminate damaged mitochondria



http://www.lookfordiagnosis.com/mesh_info.php?term=Reactive+Oxygen+Species&lang=1

evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

- mitophagy signalling pathway: enzymes PINK 1 and parkin
- Inhibitors?

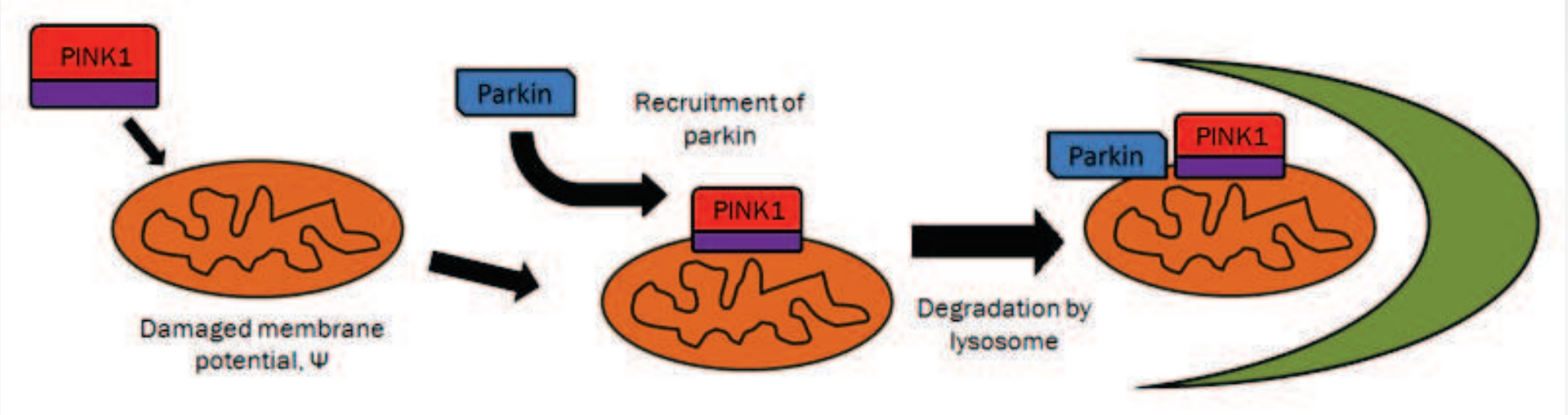


http://en.wikipedia.org/wiki/PINK1

A healthy mitochondria can import PINK1 where it is subsequently cleaved by PARL. This prevents any buildup of PINK1 and parkin is not recruited to the mitochondria.

evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

Mitophagy



of mitochondria => transfer of ubiquitin to lysin amino/acid residues of mitochondrial outer membrane proteins => degradation

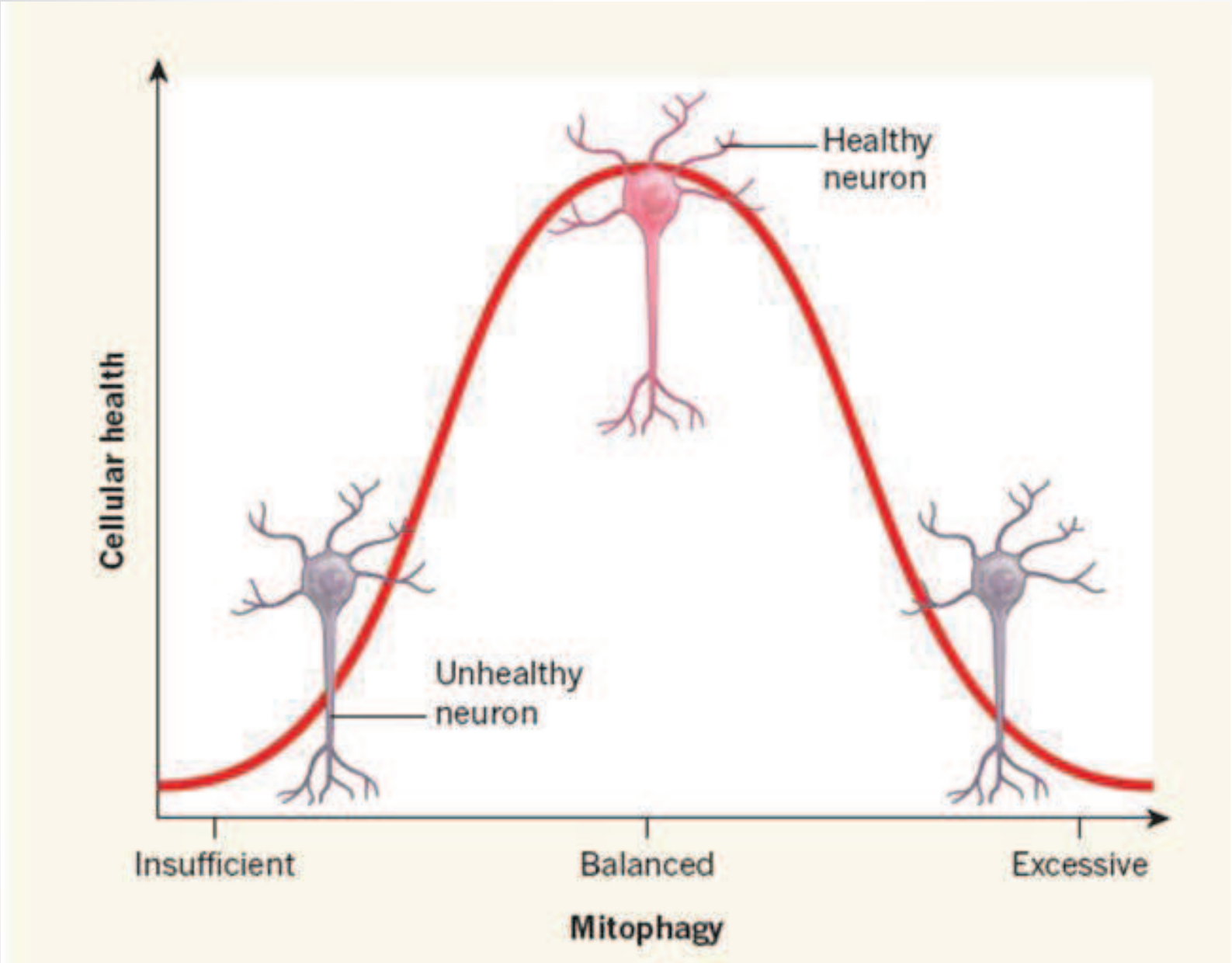
http://en.wikipedia.org/wiki/PINK1

A damaged mitochondria being recognized by PINK1. PINK1 builds up on the outer membrane of the mitochondria and recruits parkin. The PINK1/parkin pathway then designates the mitochondria for degradation by lysosomes.

esf evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

- mutations in PINK and parkin => defect in cell control => neurodegenerative disorders (Parkinson's disease)
- experimental tests: enzyme USP30 caused, that mitophagy machinery do not recognize damaged mitochondria and reverse the accumulation of ubiquitin, which is necessary
- USP30 deubiquitinate proteins, which is needed to be ubiquinate (TOM20) with some exceptions

esf evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ



Neither high or low level of mithophagy is beneficial for cells

Express both parkin and USP30 should be in balance

http://www.nature.com/nature/journal/v510/n7505/full/nature13500.html

evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

- It is the same with neurons?
- USP30 and its role is opposite to function of PINK1 and parkin
- experiments with fruit flies: defects in mitophagy were largely reversed when USP30 was removed throughout the animal
- the removal of USP30 or its inhibition can increase survival of cells in organism with Parkinson's disease

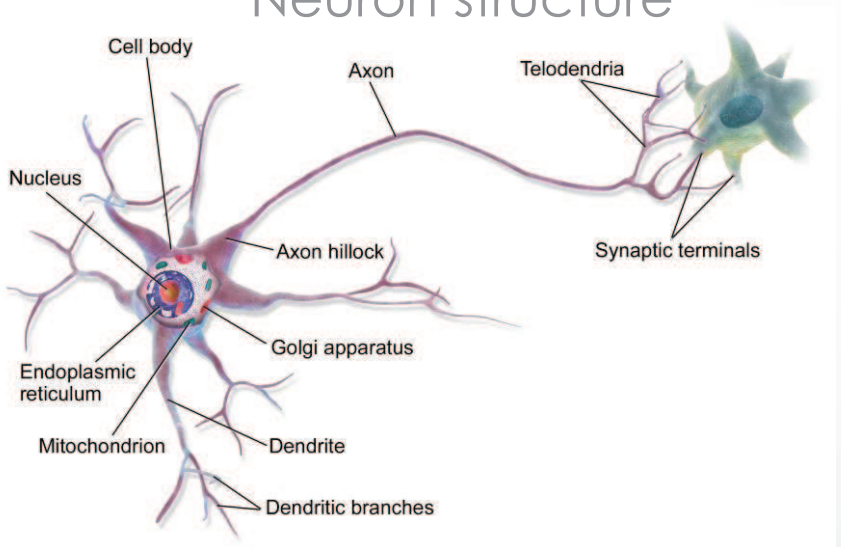
Drosophila

http://en.wikipedia.org/wiki/Drosophila

evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

- it is still unclear why dopamine-producing neurons are more sensitive to famililal mutation in the PINK1/parkin pathway that other cells in the body (levels of USP30 and parkin?)
- USP15 – similarly reverse the PINK1-parkin pathway

Neuron structure



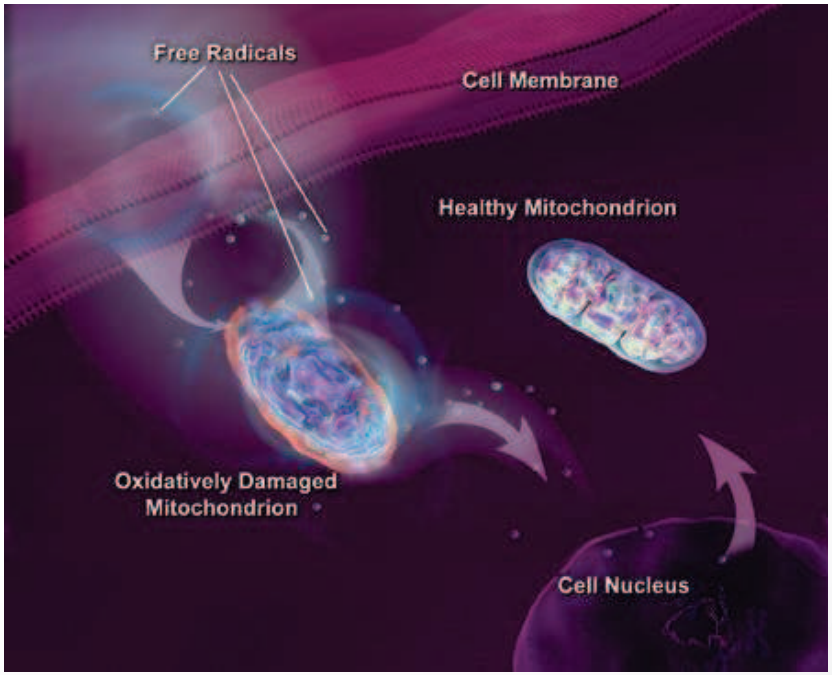
http://the-works.net/tag/basic-neuron-types

evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

What is needed to be done

- Find out what is the mechanism by witch ubiquitinated mitochondria are recognized by autophagosomes, process of degradation
- determine whether releasing the parkin brake will benefit patients

Damaged mitochondrion by free radicals



http://www.nia.nih.gov/alzheimers/scientific-images

esf evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

Thank you for your attention

evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

Propojení výuky oborů Molekulární a buněčné biologie a Ochrany a tvorby životního prostředí OPVK (CZ.1.07/2.2.00/28.0032)

Inspiration and application in the evolution of biomaterials

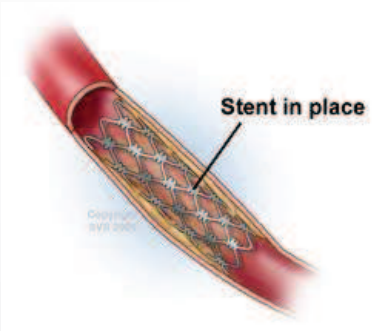
Petr Plevák

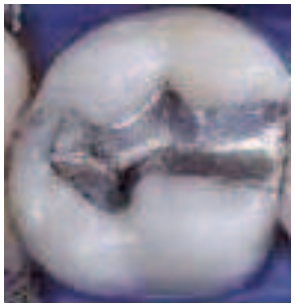
- Nathaniel Huebsch & David J. Mooney
- 426 – 432 | NATURE | VOL 462 | 26 NOVEMBER 2009

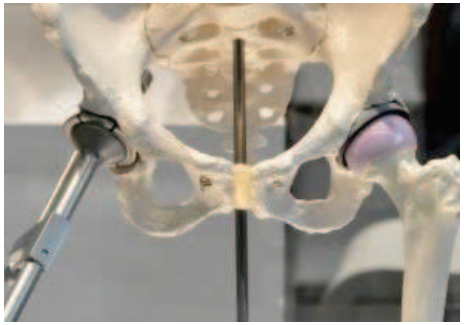
esf evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

History and growth of biomaterials

- Humankind's use of materials to repair the body dates to antiquity, when natural materials such as wood were used in an attempt to structurally replace tissues lost to disease or trauma.
- Selection of material was based on availability and the ingenuity of the making and applying the prosthetic.
- In the early part of the 20th century, naturally derived materials began to be replaced by synthetic polymers, ceramics and metal alloys, which provided better performance, increased functionality and more reproducibility e.g. (vascular stents, dental restoratives, artificial hips and contact lenses.)



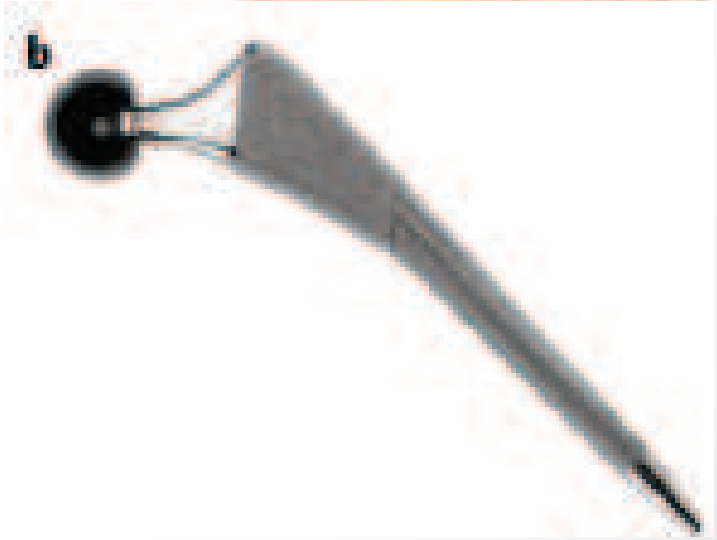




History and growth of biomaterials

a) Prosthetics fashioned from natural materials: wooden toe, cca 1065–740 bc, used as a prosthetic to replace an amputated toe and identified in an anthropological excavation of the Thebes West tombs, Egypt.

b) The synergy hip implant is an example of a state-of-the-art prosthetic device that uses synthetic materials fabricated and engineered to meet performance demands.



History and growth of biomaterials

c) Atom Stent, a metal stent from which paclitaxel is eluted into small coronary vessels to prevent restenosis (cell-mediated narrowing of the vessels).

d) The Infuse Bone Graft device, a combination product that uses both traditional prosthetic components (a steel cage) and a tissue-engineering approach (a bovine type 1 collagen sponge from which recombinant human bone morphogenetic protein 2 is eluted) to provide stability while spinal tissues are being regenerated.

esf evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

History and growth of biomaterials

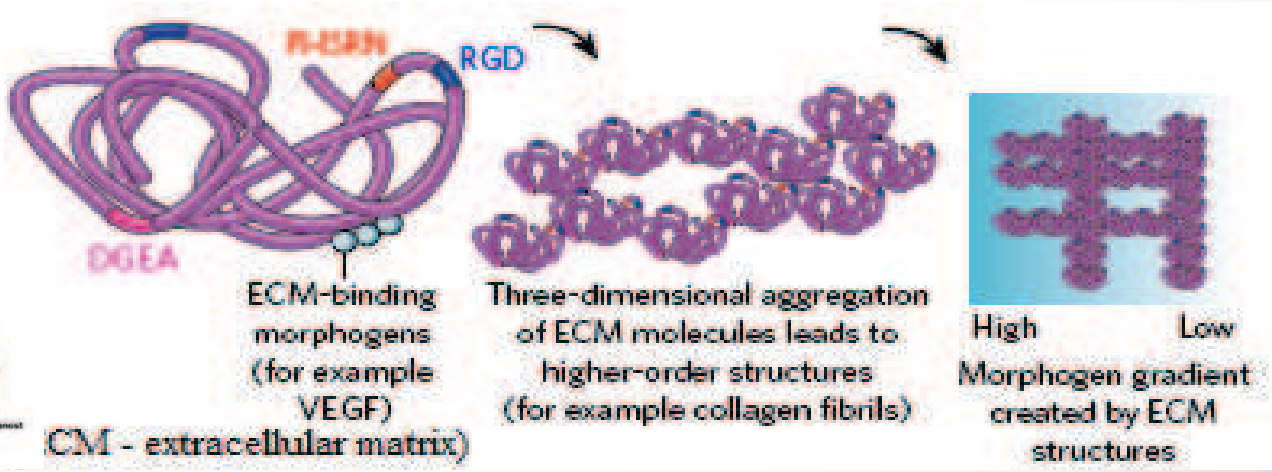
- Materials were desired to perform largely mechanical functions: to prevent biological rejection, which hampered device performance and patient health, it was preferable that they be ‘inert’ and not interact with the biology of the host organism.

- The molecular biology revolution of the 1970s and advances in genomics and proteomics, affected the ways in which biomaterials are designed and used. Specific molecules have been implicated in clinically important processes and they have been incorporated into materials as bioactive components.

- Combination products, which interface directly with cells and tissues through well-defined molecular pathways to direct biological responses, e.g. (drug-eluting vascular stents).

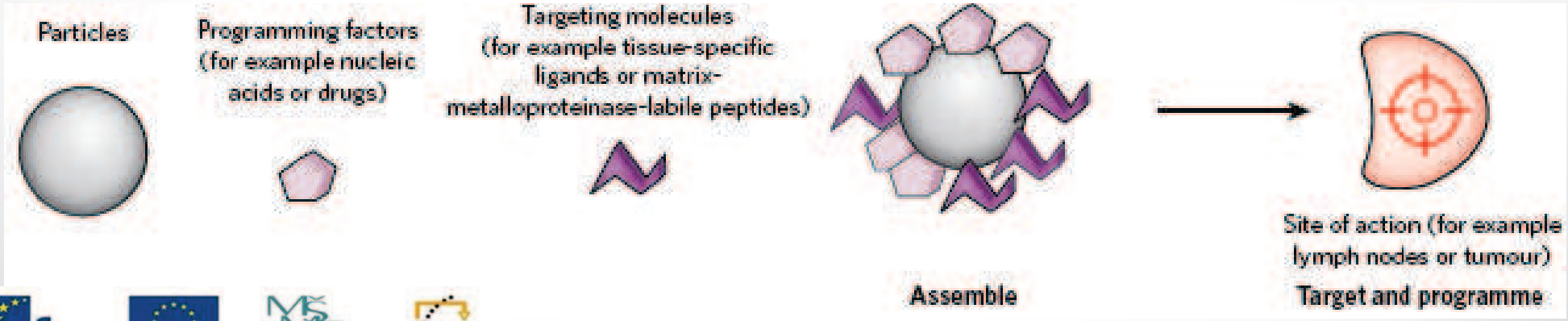
Current goals

- The life sciences are becoming equal in importance to materials science and engineering as the foundation of the field.
- Biomaterials are designed to interface with the biology of the host. This is typically done by means of binding interactions with cell surface receptors, to regulate the maintenance, regeneration or even destruction of specific tissues in the body.
- Materials can be designed to regulate host biology at a distance by controlling cell trafficking or by trafficking of the material in the body.
- Biological interactions with the host were regulated by the layer of serum proteins adsorbed nonspecifically on surfaces of synthetic materials.
- Current theme in biomaterials design is the combination of synthetics that resist nonspecific protein adsorption



Regulating biology at a distance

- Biomaterials can be designed to manipulate specific cell populations at a significant distance from the implant site.
- This can be done either by targeting the material to specific cells or anatomical locations or by controlling the trafficking of target cell populations
- Polymeric nano particles can be designed for non-invasive delivery into the body and for trafficking through the lymphatic vessels to target T cells.
- Nanoparticles are being designed to exploit the chemical and physical differences between normal and tumour-associated vasculature in order to concentrate the particles selectively within or near tumours, allowing subsequent drug-induced cell death



evropský sociální fond v ČR
EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

Physical variables

- Drug delivery from biomaterials can be manipulated using remotely applied electromagnetic fields.
- The same types of field can mediate the *in situ* assembly of scaffolds for tissue engineering.
- Ion flows caused by electromechanical stimulation can probably modulate regeneration.
- In the future, biomaterials may be engineered not only to respond to external fields and forces but also to generate these physical stimuli.

Biomaterials of the future

- One focus of research on the new generation of bioinspired materials will probably be the development of ‘smart’, multifunctional nanoparticles or implants for use in our bodies.

- These materials could target desired anatomical regions, monitor health, and report on and actively intervene in biological crises. (*ex vivo* biosensors capable of predicting disease)

Thank you for your attention