Reply to comment on "The Botulinum Neurotoxin LD_{50} Test – Problems and Solutions"

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As long as our current science is not capable of producing botox in a humane way, there can only be one conclusion: we have to discontinue the use of botox for non-life-threatening indications until our science and culture are capable of producing it in a humane way.

I can accept the argument in the comment of Dr. Pickett that a substance injected into the skin is not a classical cosmetic.

The claim of Dr. Pickett – that botox is only being given to people who are about to develop a serious depression because of their wrinkles – is unacceptable. None of my colleagues in the field of dermatology, and much less those working in walk-in botox clinics, ever seriously assess the severity of an upcoming depression caused by wrinkles. And even if they did, I am sure that most people would gladly declare to be somewhat depressed because of their wrinkles – as a matter of fact, I am too!

And yes, character does matter in this subject. We should expect from people of today's culture to decline asking mice to suffer so severely to relieve them from a few wrinkles for a short time.

The principle stating that we should not use technology we are not yet able to manage in a proper way should also apply to other fields, e.g., the REACH program. Of course it would be nice to know more about the toxicity of old chemicals. But as long as we are not able to test them in a humane manner, without harming and killing millions of animals, and in spite of the fact that we do not know how many humans will ever profit from this research, we should wait until we are able to test with animal-free methods.

Our science and culture are neither ready for botox nor for REACH.

Further information

For clarification, excerpts of the relevant texts pertaining to the above discussion from Directive 76/768/EEC and the European Pharmacopoeia are transposed here.

Council Directive of 27 July 1976 on the approximation of the laws of the Member States relating to cosmetic products (76/768/EEC)
Article 1
1. A "cosmetic product" means any substance or preparation intended for placing in contact with the various external parts of the human body (epidermis, hair system, nails, lips and external genital organs) or with the teeth and the mucous membranes of the oral cavity with a view exclusively or principally to cleaning them, perfuming them or protecting them in order to keep them in good condition, change their appearance or correct body odours.

European Pharmacopoeia 5.0, p. 1117-1119.
01/2005:2113
Botulinum Toxin Type A for Injection
Toxinum botulinicum typum A injectabile

Bulk purified toxin: Specific activity
The specific activity is confirmed in a mouse model of toxicity of by *in vivo/in vitro* methods validated with respect to the LD_{50} assay and expressed in mouse LD_{50} units per milligram of protein. Specific activity must not be less than 1×10^8 mouse LD_{50} units per milligram of protein for the 150 000 relative molecular mass neurotoxin and must not be less than 1×10^7 mouse LD_{50} units per milligram of protein for the 900 000 relative molecular mass neurotoxin complex.

Final lot: Assay
The potency of the reconstituted product is determined by an LD_{50} assay in mice or by a method validated with respect to the LD_{50} assay. The potency is expressed in terms of the LD_{50} for mice or relative to the reference preparation.
For determination of the LD_{50}, graded doses of the product are injected intraperitoneally into groups of mice and the LD_{50} is calculated by the usual statistical methods (5.3) from the mouse lethality in each group. A suitable reference preparation is assayed in parallel; the potency of the toxin is expressed relative to the reference or the value found for the reference is within suitable limits defined in terms of the assigned potency.
After validation with respect to the LD_{50} assay (reference method), the product may also be assayed by other methods that are preferable in terms of animal welfare, including 1 of the following:
1. endopeptidase assay *in vitro*;
2. *ex vivo* assay using the mouse phrenic nerve diaphragm;
3. mouse bioassay using paralysis as the endpoint.
For these other methods, the potency is calculated with respect to a suitable reference preparation calibrated in mouse LD_{50} units.
The estimated potency is not less than 80 per cent and not more than 125 per cent of the estimated potency.
The test may be repeated but when more than 1 test is performed, the results of all valid tests must be combined in the estimate of potency.