OTOTOXICITY OF ORGANIC SOLVENTS — FROM SCIENTIFIC EVIDENCE TO HEALTH POLICY

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Abstract
The scientific workshop, organized under the 6th European Framework Programme, the Marie Curie Host Fellowship for the Transfer of Knowledge “NoiseHear” Project, by the Nofer Institute of Occupational Medicine (Łódź, Poland, 15–16 November 2006), gathered world specialists in noise, chemicals, and ototoxicity, including hearing researchers, toxicologists, otolaryngologists, audiologists and occupational health physicians.
The workshop examined the evidence and the links between isolated exposure to organic solvents, combined exposure to noise and solvents, and effects on the auditory system. Its main purpose was to review the key scientific evidence to gather the necessary knowledge for developing adequate occupational health policies. This paper summarizes the workshop sessions and subsequent discussions.

Key words:
Toluene, Styrene, Mixtures, Noise, Dose-response relationship, Occupational exposure limits (OEL)

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INTRODUCTION

Workshop scope and purpose
The main objective of the workshop was to present the existing substantial evidence and highlight gaps in knowledge of the ototoxicity of organic solvents alone and in combination with noise to support the development of European legislation. Sixteen invited experts presented literature reviews and their own experience aimed at establishing standards and adequate public health policies for protecting the health of exposed workers.

The workshop comprised 5 sessions followed by discussions. Session I covered the mechanisms of noise- and organic solvent-induced hearing loss; Sessions II and III presented evidence based on animal and human studies of dose-response relationships in the ototoxicity of organic solvents alone and in combination with noise; and Session IV discussed how to suit actions to scientific evidence, e.g., setting health and safety standards for exposure to noise and organic solvents. A free paper session was also organized, providing the invited experts with the opportunity to present their recent research on noise- and organic solvent-induced hearing loss.

Seventy participants from 15 European countries, the USA and Hong Kong represented research institutions, universities, European agencies and industry. The scientific and organizing committee comprised Prof. Mariola Śliwińska-Kowalska (Project Co-ordinator), Prof. Deepak Prasher, Dr. Ewa Zamysłowska-Szmytke, and Ms. Célia Alves Rodrigues.

SUMMARY OF WORKSHOP SESSIONS AND DISCUSSIONS

Session I. Mechanisms of noise- and organic solvent-induced hearing loss
Most attention was given to the role of oxidative stress, a common mechanism of noise, drug and age-related hearing loss [1]. Recent animal studies have shown that exposure to damaging levels of noise is responsible for the increased generation of reactive oxygen species (ROS) in the cochlea, which in turn is able to induce cell death by apoptosis. This can be prevented by administering antioxidant or anti-apoptotic drugs. The results of those studies have already led to the development of antioxidant and anti-apoptotic drugs for both the prevention and treatment of noise-induced hearing loss [1].

The differences between noise- and solvent-induced hearing loss mechanisms have been discussed. The main difference concerns the exposure route, i.e., the way the agent enters the cochlea. In the solvent exposure a poisoning intrusion is emerging at the level of the inner ear [2,3]. The aromatic solvents pass through the inner ear stria vascularis in metabolized and unmetabolized forms and use the outer sulcus as the main intoxication route to reach the outer hair cells (OHCs), so that no mechanical stress is involved. Consequently their pathological mechanisms differ from those of noise. However, in both noise and solvent-induced hearing loss, the OHCs can die through necrosis or apoptosis [1,2,4]. Synergetic effects occur in animals exposed to both noise and solvents. Solvents may modify the membranous structures of OHCs making them more fragile and vulnerable to noise [3]. Thus the same acoustic energy entering the cochlea might be more harmful when accompanied by an organic solvent.

Similarities and differences between diverse solvents were evaluated. Due to their significant diversity [5,6], it has been suggested to consider them one by one when proposing safety and health measures. Following the results of animal studies it appears that styrene is probably most damaging.

Session II. Ototoxicity of organic solvents: evidence for dose-response relationships based on animal studies
Based on the literature data it has been demonstrated that the effect of solvent exposure in the rat requires a substantial organic solvent exposure over several days, before hearing impairment can be detected. The lowest concentration of styrene known to increase noise damage is 300 ppm [7] and for toluene 1100 ppm [8]. These have been the lowest concentrations tested on animals, but it cannot be ruled out that lower levels do not damage the cochlea. Styrene is regarded to be more ototoxic than toluene [9].
There are probably different toxic mechanisms involved at high exposure levels as compared to its low levels [10]. The latter could significantly contribute to the hearing effects induced by the combined exposure to noise and solvents. These observations seem to be of primary concern in humans, since combined exposures are much more common than exposures to very high levels of organic solvents alone.

The primary risk of hearing impairment attributed to organic solvent exposure may therefore be enhanced if combined with noise exposure [11–14], ethanol exposure [15,16], and other risk factors (e.g., tobacco smoking, increased blood cholesterol).

The differences in solvent ototoxicity between species have been discussed [17,18]. The rat is an animal primarily used to investigate the ototoxicity of the organic solvents. Guinea pigs do not reveal ototoxic effects of solvent exposure due to their different pulmonary uptake and liver metabolism. They do not display the effect at low exposure levels, and usually die at higher exposure levels. Taking into consideration all toxicological parameters, the rat is the best model for extrapolating the effect of solvents to humans, since both humans and rats show the same toxicological parameters [6].

The most crucial point of the discussion on this issue was how far the results of animal studies can be extrapolated to humans. A safety factor of 10 can be used when transferring knowledge acquired on rats to humans—a relatively high value that accounts for error. But the issue on which level of the risk is acceptable requires a political decision.

Session III. Ototoxicity of organic solvents: evidence for dose-response relationship based on human studies

The studies of exposure to organic solvent mixtures, styrene alone, and toluene alone in humans were summarized to derive the exposure-hearing loss relationship. Solvent mixtures are the most common kind of exposure in industry. Over the recent 22 years, fifteen studies have investigated the relationship between occupational exposure to organic solvent mixtures and hearing loss in humans. Mixtures included xylene, toluene, methyl ethyl ketone (MEK), methyl isobutyl ketone and others (ethanol, ethyl acetate, butyl acetate, ethyl benzene, thinner, cyclohexane, benzene). These studies were carried out in painters, workers of the paint and lacquer industry, dockers, petroleum refinery workers, and those exposed to jet fuel, employees of the aviation industry, chemical plants and others, characterized by varied exposures, current and life-time, to different solvents [19–25].

There is evidence that very low and short term exposures to solvents are unlikely to produce any ototoxic effect [22,23,26]. Moderate exposures to organic solvent mixture (below or around the occupational exposure limit (OEL) for each solvent), have been shown to increase the odds ratio of hearing loss [23]. Finally, in the population exposed to high concentrations of organic solvents (much above their OEL), the linear dose-response relationship was shown between exposure level (measured as lifetime exposure index), the risk of hearing loss and hearing threshold at high frequencies (in principle 8 kHz) [24]. Some studies have revealed the central effect of solvents to the auditory system, stressing that pure-tone audiometry might be insufficient to differentiate between noise and chemical ototoxic effects [27–29].

It was stressed that current exposure limits for solvents, established separately for each single chemical, are probably not effective in hearing protection when they are mixed [24].

Regarding styrene-alone-exposures that are present mainly in the glass-fibre reinforced plastic industry, only nine occupational studies carried out in different groups of workers, with different designs (mainly cross-sectional) and different outcomes are available. Of these nine studies, seven show some effects on the auditory system associated with styrene-alone-exposure. These effects are found in different audiologic tests, such as pure-tone audiometry [30,31], high frequency hearing loss [32,33], and central hearing tests [34-36]. In some studies an increased risk for hearing loss was found to be associated with exposure estimates in odds ratio calculations [30,37].

The effects found in the studies were generally insignificant and could possibly be detected only in group comparisons or risk estimation analyses. The effect was found also in the studies of very low styrene exposure [30].
the studies involving the life-time exposure calculation, this effect varied, but was rather moderate. It has been concluded that styrene is a risk factor for hearing loss. Unfortunately, based on existing studies, it is not possible to derive a dose-response relationship. Therefore, a meta-analysis is needed as the studies on large populations can yield the most evident effects.

Toluene-alone-exposures are almost exclusively found in rotogravure printing plants. There are four papers assessing the relationship between occupational exposures to toluene and hearing loss in humans. The ototoxic effect has been shown in workers exposed to high levels of the chemical (75–365 ppm) [39], while it was absent in workers exposed to toluene at the concentration below 50 ppm, using the same study protocol in the longitudinal study design [40].

Some difficulties arise if one wants to find out how to derive a dose-response relationship for a combined exposure to noise and organic solvents. Although there is a large number of investigations providing evidence for solvent and noise effects on hearing, there are also negative studies [41]. Some authors say that ototoxicity has been observed in rats, but studies in humans failed to reveal its relationship with noise, they criticize large variation in exposures, poor control groups and difficulties in exposure control. Some of this criticism is justified by the concept of conciliation proposed by Wilson who claims that even using different approaches in the study of an effect, the conclusions must be coherent.

It was stressed that even if the combined effect of noise and solvents exists at low levels of exposure, no dose-response relationship can be established before the risk groups are identified. Using pure-tone audiometry a response may underestimate the effects of combined exposures. The outcomes of the studies should be more applicable when assessing the risk of occupational hearing loss and the effect on everyday life of the exposed populations [42,43].

The tests directed towards the central nervous system processing the inflow from the auditory and vestibular nerves should be implemented to assess the central effect of solvent ototoxicity. Distorted speech testing and cortical response to frequency glides are of value as are some vestibular tests, as shown in populations exposed to solvent mixture [28] and styrene alone [34,35]. Some recommendations have been made on how to improve the efficiency of exposure measurements and risk assessment [43]. As for noise assessment, in addition to the acoustic energy of noise, its impulsiveness has to be also evaluated [44]. As leisure noise exposure seems to be increasing, it should be considered when estimating the life-time exposure. It was observed that the exposure to noise can explain only 25% of the hearing loss. The rate of explanation increases to 50% when other exposures and all known or suspected individual risk factors are considered [42]. Therefore, to obtain adequate risk assessment of noise-induced hearing loss (NIHL), a very comprehensive survey, with a large number of factors taken into account has to be carried out. Exposure to organic solvents should be one of them. This will require the use of systematic data collection and an organized database program to make the actions uniform in different countries. Moreover, it is quite likely that genetic factors should also be considered if we want to increase the rate of explained agents, which induce the occupational hearing loss.

The general conclusion was that human studies confirm the results of animal studies. The most convincing evidence for solvent ototoxicity is derived from the studies of solvent mixture exposures, where the risk for hearing loss increased with the increasing number of solvents in a given mixture [37]. Another conclusion agreed in the discussion was that styrene, the most frequently investigated solvent, shows ototoxic effects in both animals and humans.

The workshop participants were also very much interested in the mechanisms of solvent-induced hearing loss in humans and its similarities in animals. Although this problem has not yet been clarified, the human studies have already shown that solvents exert damaging effects on the peripheral and central auditory pathways. The former have been demonstrated by otoacoustic emissions, and the latter by contralateral acoustic reflex, late evoked auditory potentials, and distorted speech recognition tests.
Session IV. From scientific evidence to action: setting health and safety standards for noise and organic solvent exposure

The existing international standards on occupational noise and ototoxic substances have been presented. Exposure to harmful chemicals is one of the most debatable work hazards. In 1980, to guarantee an efficient chemical management, the International Program on Chemical Safety was launched by the International Labour Organisation, the World Health Organisation, and the Food and Agriculture Organisation as well as by the United Nations Environment Programme and Industrial Development Organisation. Organic solvents, the most hazardous chemicals as far as hearing impairment is concerned, have gradually been recognized as such and occupational exposure limits have been set to protect the exposed workers in all European countries. The use of exposure limits and the integration of workers in health surveillance programs are nowadays the most efficient international tools in controlling and assessing workplace exposures. In addition, efficient labels and international chemical safety cards play a key role in spreading the information and raising public awareness.

In the light of the findings yielded by animal studies, current solvent limits should be updated. Recent experiments have shown that a 300-ppm exposure to styrene is already ototoxic to active rats. Choosing a less severe safety factor, SF = 10, for extrapolating these data to humans it is revealed that the lowest observed adverse effect level (LOAEL) for styrene is 30 ppm, while in several countries, the occupational exposure limit for styrene is set much above this value. Based on this finding, it has already been proposed in France to decrease LOAEL from 50 to 30 ppm in addition to the required use of hearing protectors at a lower exposure [Lex<sub>8h</sub> = 80 dB (A)] [45].

The issue on how to use human health risk assessment as a policy instrument was also discussed. This is extremely important since the chemical sector is the third largest industry in Europe, with 3 million jobs depending on it and employing directly 1.7 million people.

Studies provide evidence that a change in current practice is necessary. But to achieve this, further considerations are needed in the public sector together with other stakeholders representing different interests and using their own arguments in a wider debate for either maintaining the status quo or for effecting the change for the better public interest at large. There are of course political considerations, which must keep the balance between financial implications, health and safety issues, and general public concern of the major industrial groups.

In conclusion, the scientific evidence alone is not enough to make changes in current practice. It should be accompanied by the raised public awareness and that of politicians and policy makers in terms of health and safety implications. Considerable amount of work is therefore necessary to turn the scientific evidence into public language and to describe in simple terms why the change in policy is crucial.

Session V. Recent research on noise and organic solvents-induced hearing loss

Recently published research papers concerned the following issues: the genetic factors, which might affect the degree of noise-induced hearing loss; retrocochlear and central hearing abnormalities in solvent-exposed subjects; solvent effects on the vestibular system; and potential simultaneous effects of toluene on both hearing and color vision in the longitudinal study.

CONCLUSIONS AND RECOMMENDATIONS

Animal studies

1. Several organic solvents, as exemplified by styrene and toluene, are ototoxic in rats; their exposure damages supporting and hair cells in the cochlea, accompanied by mid-frequency hearing loss. More central (or retrocochlear) hearing damage, although very likely, has not yet been clearly demonstrated in animals.

2. There are differences in mechanisms of solvent-induced and noise-induced hearing loss in the cochlea; solvents cause chemical poisoning, whereas noise causes mechanical damage. However, both mechanisms involve the enhanced generation of oxidative stress products.
3. The lowest concentration of styrene known to induce hearing damage is 300 ppm and for toluene it is 1100 ppm. Styrene is more ototoxic than toluene.

4. Synergistic effects occur in rats exposed to both noise and solvents. In combined exposures, the most important factor for inducing hearing impairment is potency of noise exposure (level, impulsiveness); concomitant exposure to organic solvents may induce impairment, whereas the exposure to noise alone may exert only little effect.

**Human studies**

1. There is increasing evidence derived from epidemiological and clinical studies that organic solvents are toxic to the auditory organ in industrial workers.

2. The most frequent occupational exposures involve solvent mixtures with xylene, toluene, and methyl ethyl ketone (MEK) as the main compounds; in some industries styrene-alone-exposure (glass-fibre reinforced plastic industry) or toluene-alone-exposure (rotogravure printing) is present.

3. There is no consensus on the lowest occupational exposure limits for solvents in relation to their effect on the auditory organ. Based on the existing data it is also difficult to derive a dose-response relationship. On the other hand, there is evidence that existing limit values might be inadequate.

4. An additive or synergistic effect occurs in the case of the combined exposure to noise and solvents, significantly increasing the odds ratio of developing hearing loss. However, solvent-induced hearing loss is dominated by that induced by noise when permanent threshold shift is assessed with pure-tone audiometry.

5. Organic solvents have detrimental effects on both peripheral and central parts of the auditory pathway. Thus, pure-tone audiogram might be insufficient to monitor this effect, and therefore, central auditory tests must be performed.

**Health policy**

1. Styrene and toluene are the most thoroughly investigated solvents and the current body of knowledge is sufficient to propose specific health and safety measures to prevent their adverse effects on the auditory organ of exposed workers.

2. Current OELs established for each single solvent might not be satisfactory for hearing protection when the solvents are mixed and/or combined with noise.

3. The LOAEL concept and the safety factor of 10 have been proposed as adequate to extrapolate findings from animals to humans and is used successfully in France in styrene exposure.

4. The relevant health surveillance measures and tests for each substance on the chemical safety cards (e.g., yearly audiograms) should be recommended.

5. A new R-phase in labeling for ototoxic endpoints should be included in the safety material of the described solvents. Similarly to toluene, the R48 denotation — Danger of serious damage to health by prolonged exposure — could be used.

6. Awareness should be raised among occupational physicians and decision makers of the fact that the current exposure limits for solvent alone and combined with noise as well as hearing conservation programs might be inadequate — the Occupational Safety and Health Administration (OSHA) could be a disseminator.

Further work of this panel of experts should be focused on: (1) providing solvent ototoxicity risk characterization and threshold limits based on currently available data in the Consensus Paper; (2) informing the EC Scientific Committee on Occupational Exposure Limits (SCOEL) on the results of their work; and (3) recommending limit setting.

**REFERENCES**


