to modify this reflex, were used as positive controls in adult and/or young Sprague-Dawley rats.

The auditory startle test consists of 3 min of acclimation to the arena followed by two consecutive stimuli performed with a 1 min interval. The first stimulus was composed of a sound at 107 dB, 10 kHz, 137 ms and the second stimulus had an increased intensity (119 dB). The test was performed on 5- and 11-week-old rats which had been treated 15 min previously with clonidine (0.4 mg/kg) by intraperitoneal injection and on 5-week old rats which had been treated with kanamycine (400 mg/kg) from post-natal days 9 to 16, also by intraperitoneal injection.

The data showed that treatment with clonidine or kanamycine decreased significantly the amplitude of the reflex and disturbed the reflex whatever the intensity of the stimulus. In addition, treatment with clonidine increased the latency of the response.

Thus, the auditory startle test system is now fully validated and is used in routine preclinical reproductive toxicology studies at CIT.

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X10
Sensory mediated behavioral effects during exposures to ethyl acrylate

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Ethyl acrylate (EA) is a substance with a pungent and annoying odor. In Germany the occupational exposure limit (OEL) is 5 ppm, with a short term exposure limit (STEL) of 10 ppm. Medians for EA odor and lateralization threshold were reported as low as 0.0066 ppb and 4.15 ppm, respectively. Chemosensory effects of EA in humans have not yet been conclusively studied. Therefore, the present study aims at investigating these effects, emphasizing also distractive odor effects.

After written informed consent was obtained, 19 healthy volunteers (10 females, 9 males) were exposed for 4 h to EA in five different conditions. These conditions included time weighted average concentrations (C_{TWA}) of 0 ppm (control condition), 2.5 ppm, and 5 ppm. For the experimental conditions both constant and varying exposures were chosen, latter ranging from 0 to 5 ppm (C_{TWA} 2.5 ppm) and 0 to 10 ppm (C_{TWA} 5 ppm). During all exposures the participants had to complete various working memory tasks. Additionally, olfactory and trigeminal perceptions were assessed before, during, and after exposure with standardized rating scales.

Olfactory and trigeminal ratings differed significantly from the control condition. For the visual-spatial working memory task the reaction times were significantly affected by EA exposure. Post hoc comparisons yielded reaction time differences about 30 ms between the 5 ppm C_{TWA} conditions and the other conditions. The error rates were elevated by 3% during these conditions. In conclusion weak behavioral effects could be substantiated for the highly annoying conditions with C_{TWA} of 5 ppm.

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X11
Sensory irritation during acute exposures to carboxylic acids—A comparison of acetic and propionic acid

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Carboxylic acids are local irritants and especially acetic and propionic acid are widely used in the working environment. For both compounds subjective signs of sensory irritation (e.g. ratings of eye initiations) have been reported among workers and experimental exposure studies. Sensitive physiological measures are available to assess this important endpoint in regulatory toxicology more objectively. The stimulation of free nerve endings of the trigeminal nerve is crucial for the elicitation of nasal and eye irritations. Intranasally, these nociceptive fibers might trigger neurogenic responses. At the cornea, nociception might increase the blinking frequency.

In two experimental exposure experiments we investigated the effect of three different concentrations of the two carboxylic acids (acetic acid: 0.5 ppm, 0.5–10 ppm, 10 ppm; propionic acid: 0.3 ppm, 0.5–10 ppm and 10 ppm) on (a) the concentration of substance P in nasal lavage fluid (NLF) and (b) the blinking frequency. In each case, 24 subjects were exposed for 4 h. Nasal lavage was conducted prior to and after the exposure. Blinking frequency was measured twice during exposure (beginning, end).

Neither for acetic acid nor for propionic acid, there was a concentration-dependent increase in blink frequency. For both the acids, there is a slight, non-significant increase of substance in the post-measure compared to the pre-measure. This increase does not follow a dose dependency.

No physiological indicators of sensory irritation were increased at concentrations as high as current OELs (German MAK). Despite the low concentration we investigated, carboxylic acids might act via ASIC receptors that are not involved in neurogenic responses.

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X12
OEL vs. DNEL or expert judgement vs. default factors—Reference values under REACh exemplified by styrene

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A crucial step in human health risk assessment under REACh legislation is the setting of derived no-effect levels (DNELs). Representing safe exposure levels for humans, they serve as reference values for possible exposure scenarios, including workplace exposure. To set DNELs, default assessment factors as given in the REACh guidance chapter R.8 are frequently applied.

For many compounds, occupational exposure limits (OELs) exist, set by scientific committees of Member States or European institutions. At this point controversy may arise, since DNELs can come

into conflict with existing OELs. In addition, different DNELs may be proposed by different registrants for a single substance.

The situation is increasingly complicated if OEL and DNEL are based on the same data set but different extrapolation procedures were used, with the OEL procedure deviating from the one recommended under REACh.

What are the consequences of different limit values for a single compound on the market? Which one should be legally binding? What if the proposed DNEL is lower than an existing OEL? Would the adherence to existing higher OELs lead to an insufficient protection of workers?

Exemplified by styrene we evaluated different ways of setting DNELs. First, default assessment factors are used, feigning ignorance on styrene's mode of action. Second, styrene-specific assessment factors are proposed based on established interspecies differences in toxicokinetics and -dynamics. The outcome of these approaches is compared to the German MAK value.

The implications of the different approaches for occupational hygiene are discussed.

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X13
Risk assessment of mixtures of mutagenic and carcinogenic chemicals: A regulatory perspective from the UK☆

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Recently the UK Government's advisory committees on carcinogenicity and mutagenicity (COC and COM, respectively) addressed the risk assessment of combined exposure to chemicals which are mutagenic and/or carcinogenic. The review focused on the use of common mechanism groups (CMG) to evaluate combined exposure risks, and the impact of potential synergistic interactions on human health.

With regards to mutagenicity, studies were identified which examined a variety of mixture types, including pollution samples, foods and hazardous wastes. The COM agreed an outline strategy for the fractionation and monitoring of the mutagenicity of chemical mixtures. Several potential hypotheses regarding synergism were identified, including the interaction between ultimate DNA reactive chemicals and those that impact on DNA structure.

With regards to the assessment of carcinogenic risk, it was concluded that the concept of dose additivity was appropriate for the evaluation of chemicals within a CMG, such as dioxins. Models used to evaluate synergistic interactions between polyaromatic hydrocarbons and heterocyclic amines were, in general, overly complex and thus extrapolation for risk assessment in humans is difficult. Assessment of the combined effects of estrogenic compounds was similarly complex. The hypothesis of a metabolic interaction to explain the apparent synergism between alcohol and tobacco smoking on cancer incidence was considered plausible. It was postulated that the combination of any mutagenic chemical with one that induces proliferation will act synergistically with regards to tumour induction and there is experimental and epidemiological evidence for this. Other stages of the carcinogenic process where chemicals could interact were also considered and will be addressed.

☆ Selected for Oral Presentation.

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X14
Health-based workplace limit values for substances affecting male reproduction

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Derivation of health-based workplace limit values (Arbeitsplatzgrenzwerte, AGW) was up to now only conducted for substances not classified as carcinogenic, mutagenic or toxic to reproduction. Since the existence of a practical threshold can also be plausibly assumed for reproductive toxicity, a comprehensive literature analysis has been performed to investigate whether the general methodology for the derivation of AGWs can be transferred qualitatively and quantitatively in relation to inter-, intraspecies and time extrapolation to the toxicological endpoints of male fertility (data presented here) and developmental toxicity (data not presented).

Toxicological effects on male reproduction are generally comparable in different species, thus justifying the use of animal studies for human risk assessment. Interspecies comparisons confirmed the common procedure of scaling according to caloric demand (basis: 50 percentiles and geometric mean). To ensure a higher level of safety, an extra factor is required in addition to scaling. Use of an additional factor for intraspecies extrapolation seems not to be justified. Reproductive toxicity is often already noticeable after four weeks if sensitive parameters such as testes histology, epididymis histology or weights of reproductive organs were examined. Time extrapolation factors derived on the basis of effects on male fertility are lower than the time extrapolation factors currently used for other endpoints. The use of additional factors for missing data has to be decided case by case.

The results of this and other research projects were further discussed by an expert working group of the German Federal Ministry of Labour and Social Affairs and put into a regulatory concept which will be presented.

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X15
Alternatives to animal testing under the EU cosmetics directive

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An amendment to the cosmetics directive of the European Union was made in 2003. It aims to phase out the use of animals in testing of cosmetic products and ingredients.

From March 11, 2009 on, the first of two deadlines bans animal use for testing toxicological endpoints after single application.