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Assessment of Human Exposure to Atrazine Through the Determination of Free Atrazine in Urine

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Atrazine (2-chloro-4-ethylamino-6-isopropylamino-1,3,5-triazine) is widely used as a selective pre-emergence herbicide on many crops; in Italy the product is extensively utilized in the culture of maize. The large quantities of atrazine used require methods for assessment of exposure to atrazine of workers engaged in its industrial production and utilization in agricolture.
Several investigations have been carried out on the metabolism of atrazine in animals.

Bakke et al. (1972), in rats orally dosed with 0.53 mg of ^{14}C-ring-labeled atrazine, found the radioactivity to be excreted mainly with the urine (80%) within 24 hr after dosing.
They identified the chemical nature of 4 of the 19 urinary metabolites of atrazine detected in urine.

Foster and Khan (1976) demonstrated the presence of unchanged atrazine and metabolites (deethylated atrazine, hydroxyatrazine and deethylated hydroxyatrazine) in excreta of hens fed with a diet fortified with 100 ppm of atrazine for seven days.

Erikson et al. (1979b), in Pittman Moore miniature pigs dosed with 0.1 g of atrazine dissolved in ethanol, found the parent compound and deethylatrazine (2-chloro-4-amino-6-isopropylamino-1,3,5-triazine) in urine within 24 hr. On the basis of this study they concluded that urinary excretion of s-triazine herbicides should be expected to occur within 24 hr mainly as the parent compound also in humans. The same Authors (1979a) described an analytical method for the separation and characterization of s-triazine herbicide residues in human urine and found levels of unchanged atrazine ranging from 0.1 to 10 ppb in the urine of a sprayer.

Bradway and Moseman (1982), in rats dosed with atrazine, simazine (2-chloro-4,6-bis(ethylamino)-1,3,5-

triazine) and propazine (2-chloro-4,6-bis (isopropyl-amino)-1,3,5-triazine) studied the urinary excretion of dealkyl-triazines (2-chloro-4-amino-6-ethylamino-1,3,5-triazine, 2-chloro-4-amino-6-isopropylamino-1,3,5-triazine, 2-chloro-4,6-diamino-1,3,5-triazine).

Studies on metabolism and excretion of atrazine in man are not available in the literature.
The present study has investigated human exposure to atrazine during its industrial production by means of assessment of ambient exposure and determination of free atrazine in urine.

MATERIALS AND METHODS

Four workers (age 34-45 years; mean 41±3), exposed to atrazine during its manufacture and packaging in a production plant, volunteered for the study.

Atrazine was determined in airborne dust of the working environment obtained by personal sampling, on skin pads according to the WHO standard method (1982), and on the skin of the hands of the workers by means of a washing procedure.

One subject (C.R., bagger) was monitored during 6 consecutive workshifts; two subjects (M.P. bagger and N.C., control-board operator) during 4 consecutive workshifts. For the fourth worker (A.A. control-board operator) only airborne exposure was measured.

Urine was collected before, during, and after exposure according to the following program:
- a 24 hr collection before the first workshift;
- all the urine voided during the monitoring period, subdivided in 8 hr fractions;
- one or more 12 hr samples after the end of the exposure period.

Each worker wore a pad on the garment in the neck area, which was used to assess skin exposure of the face. Two pads were worn underneath the suit on the skin of the trunk, one in front and one on the back. These pads were used to estimate the covered-skin exposure. Filters of the personal samplers and skin pads (λ cellulose and layered gauze backed with aluminium foil, 10x10 cm) were stored after sampling in hermetically sealed glass vials, respectively of 10 and 50 mL capacity, and treated with acetone (5 and 20 mL) in ultrasonic bath for 10 min.

Hands were washed with 200 mL of water; the washing

fluid was then treated in a separatory funnel with 25 mL methylene chloride; after shaking, the methylene chloride layer was concentrated to dryness and dissolved in 100 µL acetone.

Analysis of atrazine extracted from filters, pads, and hand washing fluid was performed by means of a Perkin-Elmer Sigma 3B GC-NPD system operating under the following conditions:
- column:pirex glass:lenght:2m; id:2 mm; phase:1.5% OV17/1.95% QF-1 on 100-120 mesh chromosorb VHP.
- temperature: oven 175°C; detector 240°C; carrier N_2 11 mL/m'-air 26 PSI-H_2 10 PSI.

For the determination of atrazine in urine, 50 mL of each urine sample, acidified with 0.5 mL of concentrated HCl, were added with 10 g NaCl and 25 mL methylene chloride; after mechanical shaking for 5 min and settling, the CH_2Cl_2 layer was transferred into a glass vial and the aqueous layer was reextracted twice. The CH_2Cl_2 phases were pooled, dehydrated by anhydrous Na_2SO_4 and, after filtration, dried by nitrogen flow and solubilized with 100 µL acetone.

Analysis was performed by means of a HRGC-MS system (Hewlett-Packard-mod 5985-B) operating on the following conditions:
-column: capillary, fused silica, lenght 25 m, id 0.3 mm; phase SE-54;
-oven temperature: initial 50°Cx2min:rate 8°C/min; final 270°C
-injector splitless: 270°C; carrier He 9.5 PSI;
-detector: selected ion monitoring;
-ionization: electron impact 70 eV.

RESULTS AND DISCUSSION

Air (TWA mg/m^3) and dermal (mg/hr) concentrations of atrazine and the urinary excretion (µg/hr) of the unchanged compound of the subject C.R. (bagger) are shown in figure 1; the monitoring period lasted for seven days (6 workshifts) for this worker. The same data concerning the subjects M.P. (bagger) and N.C. (control board operator) are reported in figure 2 and 3.
Figure 4 shows aerial exposure (mg/m^3) only and urinary excretion (µg/L) of the subject A.A. (control board operator).

Air atrazine concentrations during production and bagging were found to vary from 0.07 to 0.53 mg/m^3 (8 hr TWA); skin deposition (whole body) from 4.11 to 10.66 mg/hr. Urinary excretion of unchanged atrazine

in the exposed workers showed a pattern consistent with exposure, with maximal excretion rates of 0.1-0.3 µg/hr during the workshift and decrease to 0.01-0.04 µg/hr 12 hrs after the end of the workshift.
It is difficult to estimate the total dose received by each worker with the information generated in this study or available in the literature. The atrazine dust particles dispersed in the air at the workplace had an average dimension of 45 µm, (range 30-70 µm), thus the inhaled particles should not penetrate very deeply into the airway tree of the workers. However, the absorption rate for atrazine through the lung or the bronchial mucosa is not known, in the same way as it is not known the ability of atrazine to penetrate human skin. Experimental data on rats (CIBA-GEIGY-personal communication, 1987) indicate that atrazine penetrates the skin rather poorly; therefore it seems likely that only a minor fraction of the total amount deposited on the human skin should be taken into consideration when estimating the transcutaneously-absorbed dose. Assuming 20% and 10% absorption for inhalation and skin respectively, atrazine absorption by the workers tested in this study would be estimated to lie in the order of 3-8 mg per workshift. Free atrazine recovered in urine after exposure was in the order of magnitude of µg. This indicates that unchanged atrazine accounts for only a very minor part of the total absorbed dose. The great majority of atrazine most probably is converted into metabolites, similary to what is found in some animal species. Thus the value of the determination of free atrazine as a test for monitoring atrazine exposure is limited because of the scarce representativity of this indicator and the consequent error in calculating back the dose.
This issue seems to be confirmed by a comparative analysis of exposure and excretion results among the workers under study. Despite the fact that all the workers showed similar values in term of air and skin concentrations, one of them had urinary excretion values sensibly higher than the others.

This study showed that unchanged atrazine can be measured in human urine after industrial exposure with a pattern consistent with its absorption at the workplace. Since free atrazine in urine seems to represent only a very minor fraction of the total amount absorbed, this method can be used better for a qualitative confirmation than for a quantitative assessment of exposure. Further work is in progress to develop a monitoring procedure based on determination of atrazine metabolites in human urine.

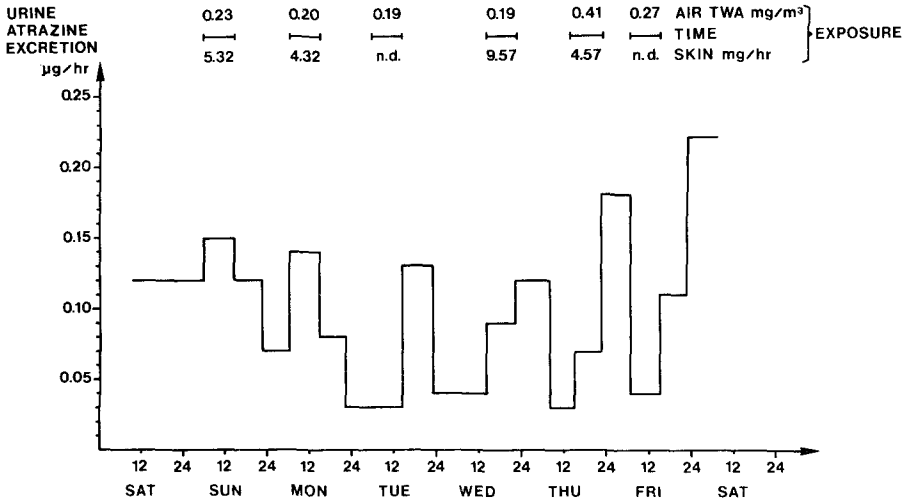

Figure 1. Inhalation and dermal exposure and urinary excretion of atrazine in a worker of a production plant (C.R., bagger)

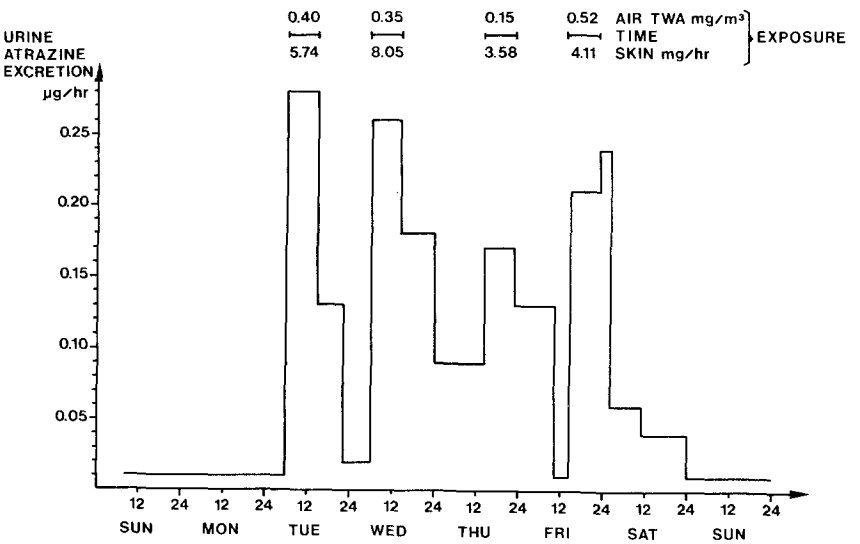

Figure 2. Inhalation and dermal exposure and urinary excretion of atrazine in a worker of a production plant (M.P., bagger).

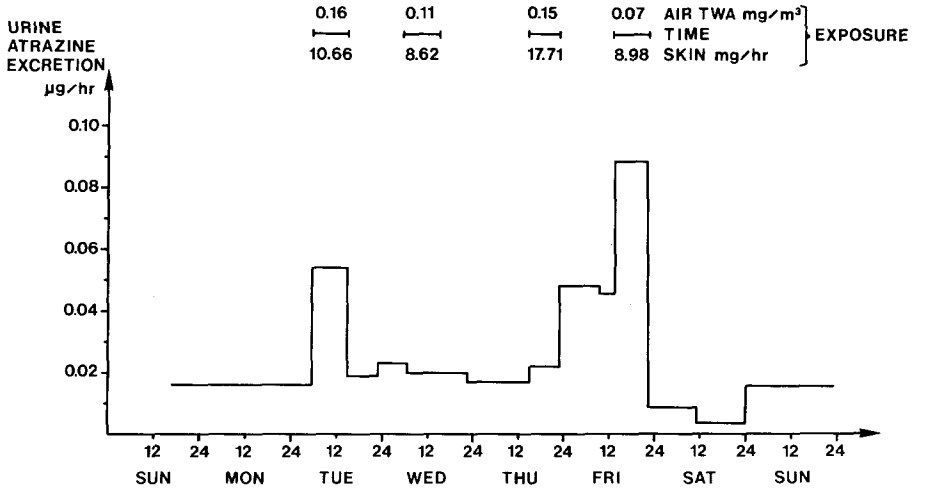

Figure 3. Inhalation and dermal exposure and urinary excretion of atrazine in a worker of a production plant (N.C., control-board operator).

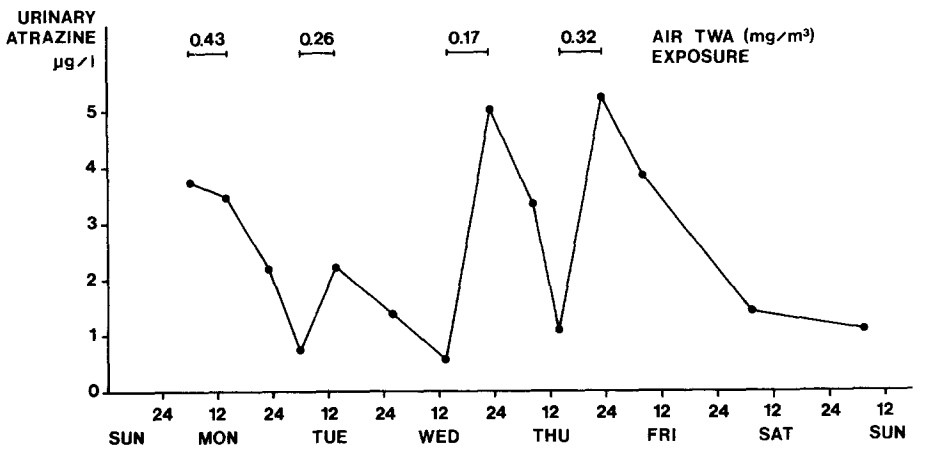

Figure 4. Inhalation exposure and urinary excretion of atrazine in a worker of a production plant (A.A., control-board operator).

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