Polish Journal of Occupational Medicine Vol. 3, No 4, 415-420, 1990

ACUTE TOXICITY OF 1,4-BUTANEDIOL IN LABORATORY ANIMALS*

RYSZARD A. JĘDRYCHOWSKI[1], JAN STETKIEWICZ[2] AND IRENA STETKIEWICZ[2]

Department of Toxicity Evaluation[1] and Department of Pathomorphology[2], The Nofer Institute of Occupational Medicine, Lodz, Poland

Key words: 1,4-butanediol, Acute oral toxicity, Acute dermal toxicity, Rat, Skin irritation, Eye irritation, Rabbit, Skin sensitization, Guinea pig

Abstract. Acute toxicity of 1,4-butanediol (BAD) was evaluated in laboratory animals. The evaluation involved acute oral and dermal toxicity in rats, dermal and ocular irritation in rabbits, and skin sensitization in guinea pigs. The oral LD_{50} values for BAD were 1.83 g/kg and 2.00 g/kg, respectively for male and female rats. The histopathological changes were observed in the liver and kidneys. No mortality was observed in female rats after dermal application of BAD at a dose of 5 g/kg. The histopathological lesions were comparable to those observed in rats after oral gavage. BAD was slightly irritant to the skin and eye of rabbits. No allergic contact dermatitis was observed in guinea pigs.

INTRODUCTION

1,4-Butanediol (BAD, CAS No. 110-63-4) is a colorless viscous liquid, miscible with water and all common organic solvents. BAD is mainly used for tetrahydrofuran and 4-butyrolactone synthesis. BAD is also utilized for production of polybutyleneterephthalate resins, thermoplastics and polyurethanes.

Among the butanediols, 1,4-BAD appears to be the most toxic (7) and was reported to produce marked hepatic damage in man and animals (3,5). The oral LD_{50} has been found to be 2.06 – 2.18 g/kg for mice, 1.52 – 1.78 g/kg for rats,

* Presented at the 6th National Congress of the Polish Association of Occupational Medicine, 20 – 22 November, 1988, Gdansk, Poland.

Address reprint requests to: R. A. Jędrychowski, Department of Toxicity Evaluation, The Nofer Institute of Occupational Medicine, P.O.Box 199, 90-950 Lodz, Poland.

1.20 g/kg for guinea pigs, and 2.53 g/kg for rabbits (5,7). Application to the eyes of rabbits showed the material to be only slightly irritating; it produced very slight conjunctival irritation but no corneal injury (7). Repeated application to the rabbits' skin, both intact and abraded, resulted in no appreciable irritation and no evidence of absorption of acutely toxic amounts (5). However, some results finding the material highly toxic on the skin were also reported (7).

The toxicological studies of BAD have been undertaken in consideration of the limited data regarding the toxicity of the chemical. The purpose of the studies described in this paper was to evaluate the acute toxicologic effects of BAD.

MATERIALS AND METHODS

BAD, obtained from the Institute of Heavy Organic Synthesis, Kędzierzyn–Koźle, Poland, was used in the study. The purity of the material, according to the information from the supplier, was above 98%.

The experimental animals were Wistar Imp:DAK rats, White Vienna and New Zealand White rabbits, and Hartley albino guinea pigs. All animals were from the Nofer Institute of Occupational Medicine husbandry. Animals were identified by unique ear number and quarantined for 7–14 days after arrival to interdepartmental animal rooms equipped with automatic light cycle timers (lights on from 6 a.m. to 6 p.m.). Relative humidity and temperature were maintained at 40–50% and 20–21°C for rats and guinea pigs, and 55–65% and 18–19°C for rabbits.

Thirty male and 30 female rats, housed in polypropylene cages (5 rats/cage) with sawdust as bedding, were used to assess acute oral toxicity. Initial body weight was 262 ± 29 g and 178 ± 15 g, respectively for males and females. Food (Murigran pelleted rodent chow, Fodder Plant, Motycz, Poland) and tap water were provided *ad libitum*. The animals were fasted from food in stainless-steel wire-mesh cages for approximately 16 hrs prior to dosing. BAD was administered by gavage at increasing doses from 1.5 to 2.5 g/kg body weight to four animals of each sex per dose. Daily observations for mortality and toxic signs were made throughout the 14-day observation period. The LD_{50} values were evaluated according to the probit method (2). Two additional groups of 5 male and 5 female rats were given BAD at the dose of 1.8 g/kg in order to assess the pathological lesions 48 hrs and 14 days after oral administration. A detailed necropsy of each animal was performed and the macroscopic appearance of internal tissues was noted. The animals selected for histopathological examination are listed in Table 1. Histological sections from adrenals, duodenum, heart, gonads (ovaries, testes), small and large intestines, kidneys, liver, lungs, pancreas, spleen, and stomach were prepared. The tissues and organs were fixed in 10% neutral buffered formalin, sectioned, stained with hematoxylin and eosin, and evaluated microscopically.

Twelve female rats, body weight 194 ± 10 g, housed individually in polypropylene cages, were used to assess acute dermal toxicity. Food and water

were provided *ad libitum*. The sides and dorsum of all rats were clipped to facilitate test material application. Test material was applied to the intact dorsum of the animals at a dose of 5.0 g/kg body weight as an undiluted liquid. The application sites were covered with gauze patches and wrapped with impervious plastic sleeves. 24 hrs after dosing, the sleeves were removed. Daily observations for mortality and toxic signs were made throughout the 14-day observation period, except for rats killed 48 hrs after application. The rats were killed and necropsied either 48 hrs or 14 days after BAD application. Similarly as in the acute oral studies selected organs were evaluated microscopically. Additionally, skin sections from the application sites were prepared. The animals selected for histopathological examination are listed in Table 1.

Table 1. Animals selected for histopathological examinations

Type of exposure	BAD-dose (mg/kg)	Time of survival	Number of animals males	females
acute	1800	48 h	4 (killed)	4 (killed)
oral	1800	14 d	3 (killed)	5 (killed)
acute	0		–	4 (killed)
dermal	5000	14 d	–	4 (killed)
(100%)	5000	14 d	–	4 (killed)

Four White Vienna rabbits, body weight 3.4 – 3.8 kg, housed individually in stainless-steel cages, were used to assess dermal irritation. Food (LSK, Fodder Plant, Motycz, Poland) and water were provided *ad libitum*. Fur from the side-areas of the trunk of all the rabbits was removed by clipping and shaving to facilitate test material application. The gauze patches (2 × 2 cm) with undiluted BAD (0.3 ml) were applied to the intact (right side) and abraded (left side) skin of animals. Adjacent areas of untreated and water-treated skin of each animal served as a control for the test. The patches were covered with plastic foil and protected by means of a suitable occlusive dressing for the 24 hrs exposure period. Observations for dermal irritation were made 1, 24, 48 and 72 hrs after patch removal. Dermal evaluation utilized the scoring system of Draize et al. (1). Dermal irritancy ratings were determined using the classification system as follows: slightly irritating < 2, moderately irritating $2-5$, severely irritating > 5 (8). The rabbits were used to assess short-term dermal irritation. The same internal areas of the right ears of rabbits were painted with either 100% or 50% BAD water solution for 10 consecutive days including weekend. The left ear of each rabbit painted with water served as a control for the test. Observations for dermal irritation were made the day after painting.

Ocular irritation studies were conducted in New Zealand White rabbits which were housed as described above. BAD was administered as a single dose of 0.1 ml, which was placed in the conjunctival sac of the right eye of four

rabbits. The unexposed eye of each rabbit served as a concurrent control. The examination for signs of ocular irritation was made at intervals of 1, 24, 48 and 72 hrs postdosing according to the method of Draize et al. (1). A Draize score was computed at each observation time by averaging the total scores of all rabbits tested. For classification the ocular irritation index was computed by averaging Draize scores at 1, 24, 48 and 72 hrs. Eye irritancy ratings were determined using classification system as follows: no reaction 0, slightly irritating 1–16, moderately irritating 17–35, severaly irritating 36–75, extremely irritating 75–110 (8).

The guinea pig maximization test of Magnusson and Kligman (6) was used to assess allergic contact dermatitis. Thirty albino male and female Hartley guinea pigs, body weight 358 ± 36 g, were kept in stainless-steel cages (3–4 animals per cage), and had free access to food (LSK, Fodder Plant, Motycz, Poland) and tap water with the addition of vitamin C (400 mg/l). The animals were randomly divided into sensitized animals (20) or control group animals (10). In induction procedure, on the basis of preliminary studies, BAD was applied at a concentration of 10% (intradermal injections) and 30% (topical application). The challenge procedure was done with 10% and 30% BAD.

RESULTS

Estimated LD_{50} values following single oral adminstration of BAD were 1.83 g/kg and 2.00 g/kg, respectively for male and female rats. Low and high 95% fiducial limits were in males 1.70 g/kg and 1.98 g/kg, and in females 1.80 g/kg and 2.22 g/kg, respectively. Deaths occurred within 48 hrs after oral dosing. The most common signs of toxicity observed in both sexes were irregular decreased respiration and catalepsy. Gross pathological findings in animals that died included a fluid-filled gastrointestinal tract and congestion of internal organs. Histopathological changes in rats killed 48 hrs after administration of BAD at a low-lethal dose of 1.8 g/kg were found in the liver and kidneys. Extensive vacuolar degeneration of the hepatic parenchyma was found in the liver of all rats. In one male rat marked periportal fatty changes were observed. The changes in kidneys were characterized by the presence of hyaline or granular casts and clusters of desquamated cells, mainly in the renal tubule lumen of the subcortical zone and the outer medulla. The tubular epithelium in place of the casts occurring was flattened. Groups of tubules with regeneration signs and interstitial infiltration with mononuclear cells were also observed. Periportal vacuolization of the hepatocytes cytoplasm and the cells in mitosis was the most prominent feature in the liver after 14 days. At that time, in the kidneys of 3/3 males and 2/5 females hyaline casts were present, also traits of single tubules regeneration and dispersed interstitial infiltration with lymphocytes.

There was no mortality in animals dermally exposed to BAD at a dose of 5 g/kg. The histopathological changes, both at 48 hrs and 14 days thereafter, were limited to the skin and liver. After 48 hrs dermal lesions were noted in 2/4

rats and were characterized by segmentary acanthosis, single microcrusts with granulocytes infiltrations, slight collagen edema and mononuclear cell infiltrations in the hypodermis. At the same time, extensive vacuolar degeneration of the hepatocyte cytoplasm was the most prominent feature in the liver of all animals. In all rats after 14 days there were observed small single desquamating crusts on the skin surface and small focal granulocyte infiltrations in the superficial layers of the epidermis. In the liver the most prominent feature was moderate to marked periportal vacuolization of the hepatocytes cytoplasm.

No dermal reaction were noted in rabbits with intact and abraded skin. There was no skin reaction following short-term dermal exposure to 50% BAD. Only after 10 days a minimal reddening of the ear skin was observed in 2/4 rabbits treated with undiluted BAD.

Slight reddening of the conjunctives and small amounts of discharge were observed in all rabbits 1 hr after ocular application of BAD. The changes diminished after 24 and 48 hrs. No abnormalities were observed thereafter.

No allergic contact dermatitis was observed in guinea pigs sensitized with BAD.

DISCUSSION

The oral LD_{50} value for the rat indicates that BAD may be classified (4) as a slightly toxic substance. This is in agreement with previously published information (5, 7). In acute poisonings BAD is a moderately hepatotoxic and nephrotoxic substance. The liver and kidneys changes caused by a single oral administration of sublethal doses of BAD are reversible, and the changes visible after 14 days can be interpreted as a reaction to the lesions in the past.

BAD is slightly irritating to the skin and eye. However, it is easily absorbed through the skin inducing pathological lesions similar to those observed after acute oral doses.

Our studies indicate that BAD has no skin sensitizing potential in the quinea pig.

ACKNOWLEDGEMENTS

This study was supported by the CPBR project 11.11.1.11. The authors are grateful for the technical assistance of Janina Kłosek and Anna Latusek.

REFERENCES

1. Draize J H, Woodard G, Calvery H O. Methods for the study of irritation and toxicity of substances applied topically to the skin and mucous membranes. J Pharm Exp Ther 82, 377–390, 1944

2. Finney D J. Probit Analysis, 3rd ed, Cambridge Univ. Press, Cambridge, 1971.

3. Hinrichs A, Kopf R, Loeser A. Zur toxikologie des 1,4-butanediol. Pharmazie 3, 110–112, 1948.

4. Hodge H C, Sterner J H. Tabulation of toxicity classes. Amer Ind Hyd Assoc Quarterly 10, 93–96, 1949.

5. Knyshova S P. Biological effect and hygienic significance of 1,4-butynediol and 1,4-butanediol. Gig Sanit 33, 37–41, 1968

6. Magnusson B, Kligman A M. Allergic contact dermatitis in the guinea pig. Thomas Publ., Springfield, 1970.

7. Rowe V K, Wolf M A. Butanediols. In: Patty's Industrial Hygiene and Toxicology. eds C D Clayton, F E Clayton, Wiley, New York, 1982.

8. Sokal J A, Knapek R, Majka J, Barański B, Jędrychowski R. Scope and methods in toxicity testing of industrial chemicals. Inst Occup Med Publ Vol. 6, Lodz, 1990 (in Polish).

Received for publication: 16th October, 1990.
Accepted for publication: 15th November, 1990.