

CODING FORMS FOR SRC INDEXING

Microfiche No.	OTS0559911-1		
New Doc ID	89010000044	Old Doc ID	8EHQ-1100-14711
Date Produced	11/20/00	Date Received	11/22/00
		TSCA Section	8E
Submitting Organization	AN GROUP INC		
Contractor			
Document Title	SUPPORT: PUBLICATION: EFFECTS OF ACRYLONITRILE ON THE IMMUNE FUNCTION IN MICE (JOURNAL OF SANITARY TOXICOLOGY. 4(4): 245. 1990). W/COVER LETTER DATED 11/20/2000 (ENGL TRANSLTN)		
Chemical Caegory	ACRYLONITRILE		

8EHQ-100-14711 41522

AN GROUP

November 20, 2000

Document Processing Center (7407)
Attention: 8(e) Coordinator
Office of Pollution Prevention and Toxics
U. S. Environmental Protection Agency
401 M Street, S.W.
Washington, Dc 20460

RECEIVED
DPPT/CBIC
2000 NOV 22 PM 1:00

CONFIDENTIAL NO CBI

Dear 8(e) Coordinator;

RE: File [8EHQ-400-14711]

As was indicated in our previous letters, the AN Group, Inc. is a U.S. trade association representing producers and users of acrylonitrile, and has submitted a number of Chinese surveys and studies on acrylonitrile to the EPA 8(e) coordinator.

We have recently received an English translation of one additional Chinese toxicology study on acrylonitrile. It was published in the Journal of Sanitary Toxicology Vol. 4, Issue 4, 1990. Neither the AN Group or any individual member of the AN Group has made a determination as to whether a significant risk of injury to health or the environment is actually presented by the findings. However, the information in the translated study may meet EPA's guidance for reporting under TSCA 8(e) and accordingly the AN Group is submitting this material for inclusion in file/docket # [8EHQ-0400-14711].

RECEIVED
DPPT/NCIC
2000 NOV 29 PM 3:01

The US acrylonitrile producers are committed to pursuing a better understanding of the quality and meaning of this study as well as previous submissions, and to incorporating any scientifically sound information into management of acrylonitrile health risks. Please contact me if you have any questions or comments about these reports.

Sincerely,

John F. Murray
John F. Murray, CAE
Executive Director



8EHQ-00-14711



Effects of Acrylonitrile on the Immune Function in Mice

Xie Guoliang, Sanitation and Anti-epidemic Station, Inner Mongolia Autonomous Region
Wang Yingcong, Labor Health Division, Lanzhou Medical College

Acrylonitrile (AN) is an important organic synthetic monomer, which is widely used in the manufacturing of nitril fiber, butadiene-acrylonitrile rubber and synthetic resin. Many studies have been done abroad on its carcinogenic, teratogenic, mutagenic and hepatotoxic effects (1). However, very few published studies have dealt with its effects on the immune function. This paper is aimed at this particular area and hopefully will provide useful reference for the assessment of acrylonitrile-related occupational hazards.

Materials and Methods

I) Materials

The animals tested were male, healthy mice of Kunming breed provided by the Animal House of Lanzhou Medical College. Their body weight averaged from 18 to 22 g.

AN was provided by Lanzhou Chemical Industry Corp. According to gas chromatography measurement, its purity was and above 99.7%.

II) Methods

The mice were divided at random into one comparison group and three contaminated groups, each group consisting of 10 mice. The mice were exposed to AN through quiet inhalation. The AN concentration ratio of the three contaminated groups were 60, 90 and 120 mg/m³ respectively. The mice inhaled AN four times a day, and the exposure period went on from one to three weeks.

III) Observation Index

1. Weighing immune organs: Mice were killed 21 days after exposure. The thymus gland and spleen were taken out and weighed. Thymus and spleen indexes were used for all measurement.
2. Abdominal macrophage (M ϕ) phagocytic function test: 14 days after exposure, M ϕ phagocytic ratio and phagocytic index were determined with the methods described in Document (2).
3. Test of humoral immune function: 15 days after exposure, 0.5 ml of sheep red blood cell (SRBC) suspension was injected into each mouse's abdominal cavity. Its eyeballs were extracted to draw blood samples and make blood serum; its spleen was extracted to prepare splenic lymphocyte suspension. Function of the primary antibody-forming cells

(IgM, AFC) in the spleen was determined (3); Anti-SRBC anti-body level in the blood serum was checked. All results are shown in OD value.

4. Cellular immune function test: Blood was drawn and smeared from the mouse's tail 21 days after exposure. Acid α -naphthyl acetate esterase (ANAE) was stained using the methods mentioned in Document (5). For each piece, 100 lymphocytes were observed under oil immersion lens to count the ANAE active and positive rates. Lymphocyte transformation function was determined using the methods described by Mosmann, Xue Bin *et. al.*

Results and Discussion

I) The effects of AN on the mouse's body weight growth: Effects on the mouse's weight growth is determined by comparing its body weight 21 days after initial contamination with its pre-exposure body weight. The results, which is shown in Table 1, demonstrate that the mice in each poisoned group gained body weight much slower than those in the comparison group. The difference is significant.

Table 1 The Effects of AN on the Mouse's Body Weight Growth

Group Concentration (mg/m ³)	Number of Animals (n)	Absolute Weight Gain (g) ($\bar{X} \pm SD$)	P Value
0	10	6.34 \pm 2.34	
60	10	2.73 \pm 1.29	<0.01
90	8	2.49 \pm 1.85	<0.01
120	8	1.20 \pm 1.67	<0.01

II) The effects of AN on the mouse's immune organs: Both the absolute and relative weight of the immune organs of each mouse in every contaminated group dropped significantly, and the dose-response correlation can be established (See Table 2). Such a result demonstrates that AN has an evident toxic effect on the immune organs. The change in the weight of spleen and thymus gland indicate a possible change in the mouse's humoral and cellular immune functions.

III) The effects of AN on the mouse's M ϕ phagocytic function: 14 days after the three groups had been AN-contaminated with their respective concentration level, both the M ϕ phagocytic ratio and phagocytic index were found to be conspicuously lower than that in the comparison group. The difference is highly significant, and the dose-response correlation can be established (See Table 3). The result indicates that AN acts as an inhibitor against the nonspecific immune function of the organism.

IV) The effects of AN on humoral immune function in mice: The IgM level in the blood serum from mice in the contaminated groups turned out to be much lower than that in the comparison group, and the decrease correlates with the increased toxic dosage. The splenic IgMAFC function in the 60mg/m³ group shows a decrease but bears no statistical significance. However, the decrease becomes obvious in the 90 and 120 mg/m³ groups (See Table 4). Such a result demonstrate that AN inhibits humoral immune function and its inhibition capacity may correlate with its concentration ratio. If highly concentrated, AN may act as a direct inhibitor against the antibody-forming lymphocyte, thus blocking the formation of antibodies; if thinly concentrated, AN can not have a direct impact on the antibody-forming lymphocyte, instead, it might act as an inhibitor against complement activation or it may decrease complement formation. In this case, even though the formation of antibodies may not be affected, the immune hemolytic reaction is decreased, resulting in a relative decrease of IgM level in blood serum when using hemolytic testing methods.

V) The effects of AN on cellular immune function: The active and positive rates of ANAE in the mice in each contaminated group are all conspicuously lower those of the comparison group, and the dose-response correlation can be established. The lymphocyte transformation rates in all the contaminated groups are also notably lower than that in the comparison group (See Table 5). The results demonstrate that AN has an obvious inhibition effect on the cellular immune function.

Table 2 The Effects of AN on the Weight of the Mouse's Immune Organs

Group Concentration (mg/m ³)	Number of Spleen Animals (n)	Spleen		Thymus Gland	
		Absolute Weight	Index	Absolute Weight	Index
0	10	246.5±56.4	92.4±17.6	99.5±31.8	37.6±10.5
60	10	175.2±56.8 **	64.5±24.3* *	73.5±42.8* *	29.2±15.9* *
90	8	132.5±35.8 *	54.2±14.2* *	44.4±12.1* *	18.8±9.7**
120	8	119.4±31.7 **	52.9±14.1* *	40.6±17.2* *	18.0±1.7**

Note: Organ Index=organ weight(mg)/body weight(g)x10 compared with the uncontaminated group *p<0.05, **p<0.01

Table 3 The Effects of AN on the Mouse's Abdominal Macrophage Phagocytic Function (X±SD)

Group Concentration (mg/m ³)	Number of Animals	Phagocytic Rate (%)	Phagocytic Index
0	9	31.50±4.4	0.53±0.15
60	8	21.24±4.1**	0.34±0.08**
90	8	18.01±3.4**	0.23±0.05**
120	8	15.11±3.1**	0.18±0.04**

* Notes: Same as in Table 2

Table 4 The Effects of AN on the Mouse's Humoral Immune Function (X±SD)

Group Concentration (mg/m ³)	Number of Animals	Splenic IgMAFC	Blood Serum IgM
0	10	0.498±0.12	0.730±0.14
60	10	0.477±0.10	0.47±0.11**
90	10	0.202±0.11**	0.296±0.13**
120	10	0.187±0.09**	0.280±0.11**

* Notes: Same as in Table 2

Table 5 Effects of AN on ANAE Active and Positive Rates and Splenic Lymphocyte Transformation Function (X±SD)

Group Concentration (mg/m ³)	ANAE Active and Positive Rates		Splenic Lymphocyte Transformation Function	
	Number of Animals	Positive Rate (%)	Number of Animals	OD Value
0	10	58.70±4.11	10	0.403±0.04

60	9	41.11±6.12**	9	0.323±0.06**
90	8	31.25±5.90**	8	0.288±0.03**
120	8	30.38±8.18**	7	0.211±0.07**

* Notes: Same as in Table 2

References

- 1, Hashimoto Waro (?Japanese name), *Occupational Medical Science*, 1980, 22(5), 327
- 2, Zhang Yunfen, et. al., *Journal of Beijing Medical College*, 1979, (2), 114
- 3, Simpson MA, et. al., *J Immunol Methods*, 1979, 21:159
- 4, Xu Xueying, et. al., *Pharmacy*, 1979, 14 (7), 445
- 5, Mueller J. *Eur J Zmmunol*, 1975, 21:152
- 6, Mosmann, T. *J Immunol Methods*, 1983, 65:55
- 7, Xue Bin, et. al., *Journal of Sanitary Toxicology*, 1988, 2 (1), 61