**MICHIGAN DEPARTMENT OF ENVIRONMENTAL QUALITY**

INTEROFFICE COMMUNICATION

December 5, 2003

TO: File for N-vinylpyrrolidinone-2 (88-12-0)

FROM: Marco Bianchi

SUBJECT: Initial Risk Screening Level

The initial risk screening level (IRSL) for N-vinylpyrrolidinone-2 is 0.04 $\mu g/m^3$ based on an annual averaging time.

The following references or databases were searched to identify data to determine the IRSL: IRIS, HEAST, NTP Management Status Report on-line, RTECS, EPBCCD, EPB library, CAS-online, NLM-online, IARC on-line, NIOSH Pocket Guide, and ACGIH Guide.

N-Vinylpyrrolidinone-2 (NVP) is an industrial raw material used in the pharmaceuticals, and cosmetics industry as a monomer in the production of homopolymers such as water-soluble and insoluble forms of polyvinylpyrrolidone. Prior toxicity tests have shown three target sites for NVP toxicity; the skin and mucous membranes, the liver, and certain peripheral blood parameters. (Greim, 1994; VCI, 1992).

Klimisch, et al (1997), conducted a series of 11 toxicity studies from 1983-1987 aimed at defining the toxicity profile of NVP under conditions of repeated exposures. Nine of the studies were by the inhalation route of exposure, while the remaining two were oral studies. The objective of the inhalation studies was to provide guidance for the selection of exposure concentrations suitable for long-term toxicity/carcinogenicity testing in rats. The nine inhalation studies involved exposure of rats (two different strains), mice or hamsters to NVP at a concentration of up to 120 ppm (545.5 $mg/m^3$), 6 hrs/day, 5 days/wk over a period of 1 week to 12 months. The two oral studies involved exposure of rats to NVP through the drinking water or by gavage at dose levels of up to 100 mg/kg body weight/day. Reduced body weight gain was seen in rats exposed by inhalation to 5 ppm or more for 3 months and in mice and hamsters exposed to 45 ppm for only 1 day. Effects were seen on hematological (reduced hemoglobin, erythrocyte count, hematocrit) and clinical chemistry parameters (raised g-glutamyltransferase activity and decreases in plasmaprotein), liver weight increase and liver lesions (centrilobular single-cell necrosis and foci of hepatocellular alteration) in rats and mice but not hamsters. Rats exposed to 40 mg/kg/day NVP or more for 3 months by gavage developed similar liver changes. Atrophy of olfactory epithelium and hyperplasia of nasal respiratory epithelium was seen

in rats exposed by inhalation to 5 ppm (22.7 mg/m$^3$) NVP for 7 weeks but not in response to 1 ppm for 13 weeks (no-observed-adverse-effect-level, NOAEL). These studies indicated that the upper respiratory tract and the liver are the main targets for NVP toxicity.

In a companion study by Klimisch, et al (1997) published at the same time as the toxicity investigation listed above; long-term inhalation toxicity/carcinogenicity of NVP was examined in two rat studies. In the first study, designated as study A, Sprague-Dawley rats were exposed to 0, 5, 10, or 20 ppm NVP for 6hrs/day, 5 days/wk for 24 months. satellite groups were sacrificed after 3, 12, or 24 months. Survival was unaffected, but reduced body weight gain, hemotoxicity, effects on clinical chemistry parameters indicative of hepatotoxicity, increased liver weight, hepatocellular carcinomas, necrosis reparative hyperplasia, adenomas and adenocarcinomas of the nasal cavity, and squamous cell carcinoma of the larynx were seen. In study B, female Sprague-Dawley rats were exposed to 0 or 45 ppm NVP for 3 months and sacrificed at 3 or 12 and 24 months post-exposure. The effect of NVP on body weight evident at 3 months disappeared before 1 year, but effects on liver pathology persisted throughout the subsequent 21-month exposure-free period, and a few liver tumors were seen at 2 years. NVP gave negative results in a battery of *in vitro* and *in vivo* genotoxicity tests with and without activation as appropriate, suggesting that the tumors that arose were manifestations of a non-genotoxic mechanism. These tests included, the Ames test, mouse lymphoma assay, unscheduled DNA synthesis, cell transformation test, Drosophilia, and *in vivo* studies using differently radiolabeled forms of NVP.

Survival was not effected throughout the study from NVP exposure. However, inhalation of NVP had an influence on body weight gain being evident as a dose-related retardation of growth. Male rats were slightly more sensitive than females. This decrease of the body weight gain was observed only during the first third of the total exposure period of 104 weeks. After that part of the exposure period the animals probably adapted to the NVP exposure; body weight was no longer decreased to a biologically meaningful extent. After a 3-month exposure the highest body weight decrease of 11% in male rats at 20 ppm was slightly above the 10% level recommended maximum tolerated dose.

The changes in clinicochemical parameters (decreased plasma-protein concentrations and increased reduced glutathione levels and g-glutamyltransferase activities in liver homogenate) revealed liver toxicity. The effects are slightly more pronounced after a 3-month exposure than after a 12-month exposure and were particularly seen in the female animals. In addition, after 12 months of exposure, an increase in cholesterol level (20 ppm females) and a slight reduction in alanine aminotransferase activity (10 ppm and 20 ppm females) was probably also caused by liver toxicity. Moreover, the changes in the red blood cell count of the female animals in the intermediate and higher concentration groups, which were seen only after a longer exposure period (after 12- and 24-month exposure), were indications of an ongoing anemic process. Since there are no concomitant findings which can explain the adverse effects on the red blood cells, it is difficult to interpret the anemic process in terms of its likely pathogenesis. However, it is

possible that there is a relationship between the changes in the red blood cell count and the decreased protein concentrations in the plasma.

The most relevant non-neoplastic findings observed in rats after exposure to the inhalation of NVP vapor were manifestations of systemic toxicity in the liver and of local toxicity in the respiratory tract (nasal mucosa and larynx). The latter were indicative of an irritating potential of NVP and were found in animals at all concentrations. The following changes were observed.

Degenerative lesions of the hepatocytes were the major treatment-related non-neoplastic findings in histopathology which may be related to the effects observed in clinical chemistry. Three treatment-related non-neoplastic lesions were observed in males and females; spongiosis hepatic, foci of cellular alteration, and focal hepatocellular hyperplasia. Spongiosis hepatic, a degenerative lesion of hepatocytes, occurred at all study intervals with increasing incidences in male or female rats. Foci of cellular alteration of various types occurred in all sacrifice groups of the study more often or even exclusively in exposed animals. After a treatment period of 3 or 12 months, the predominant cell type within the foci was the clear cell type, expressing a degenerative cell lesion. Eosinophilic foci of cellular alteration appeared to be increased in a dose-related fashion in male and female rats. They also occurred in control animals of the main groups, however, in a very low incidence. Foci hepatocellular hyperplasia occurred more often in exposed animals from 12 months onwards in one or both sexes than in the concurrent control groups.

In the nasal cavity, treatment-related non-neoplastic lesions consisted of different types of focal hyperplasia including hyperplasia of basal cells in areas lined by respiratory or olfactory epithelium, atrophy of the olfactory epithelium, squamous cell metaplasia and inflammation. Lesions of this kind were observed in all groups, though a clear dose-response relationship was not evident. In the larynx, focal epithelial hyperplasia occurred only in exposed animals. Inflammation was observed in treated and untreated males, whereas none of the control females exhibited focal laryngeal inflammation.

In summary, non-neoplastic toxic effects were evident in a dose-dependent manner in the liver and in the respiratory tract (nasal mucosa, larynx) in all NVP-exposed groups and these indicate that chronic toxicity persists throughout the exposure period of 2 years.

The liver, the nasal cavity, and the larynx were found to be target organs for the development of neoplasia. Hepatocellular carcinomas were observed in male and female rats after 24-month exposure to NVP vapor. After an exposure period of 18 months followed by a 6-month recovery period (interim sacrifice groups), the incidence of animals with hepatocellular carcinomas was significantly less than that in animals receiving NVP continuously for 2 years. After a treatment period of 12-months (interim sacrifice groups), a hepatocellular adenoma was reported in a high dose male. No hepatocellular neoplasms were seen in animals exposed for 3 months (interim sacrifice groups).

The study investigators thought it noteworthy, that no lesion considered a benign hepatocellular neoplasm was seen in any main group rat. In fact, the only lesion of this kind in the whole study was in a high dose male interim group. The observation derived from the interim subgroups indicated that inhalation of NVP resulted in hepatotoxicity. The development of hepatocellular neoplasms may be related to these findings of toxicity in relation to pathogenesis. In all rats (except of one high-dose female) that had a hepatocellular carcinoma of the liver, there was evidence of non-neoplastic liver pathology. In no rat that developed such a carcinoma was the liver free from evidence of toxicity.

In the nasal cavity, adenomas and adenocarcinomas were noted as treatment-related findings. The morphology of adenomas and adenocarcinomas is totally different. The adenomas arise from respiratory epithelium or the underlying submucosal glands of the anterior parts of the nasal cavity, whereas adenocarcinomas seem to arise from the olfactory epithelium or from underlying submucosal glands in the posterior parts. From these observations the adenomas and adenocarcinomas are interpreted as separate entities which must be evaluated separately.

Adenomas of the nasal cavity were first seen in animals that had been exposed for 12 months. The incidence was higher after 18 months of exposure and a 6-month recovery period and after 24 months of exposure. No adenomas were seen in the nasal cavity of control animals of either sex at any time during the study. Adenocarcinomas were observed after 24 months of exposure, only. They occurred more often in males than in females. In females, adenocarcinomas were only observed in high dose rats, whereas in males this type of tumor was observed in the mid- and high-dose groups. In most of the rats that had either an adenoma or an adenocarcinoma of the nasal cavity, there was evidence of inflammation and/or hyperplasia, respectively. Chronic inflammation and associated hyperplasia might have been involved in the pathogenesis of the nasal tumors. There was no clear difference between interim exposed rats and the main groups in the occurrence of adenomas except that some main group animals exposed to the lowest concentration of NVP developed adenomas.

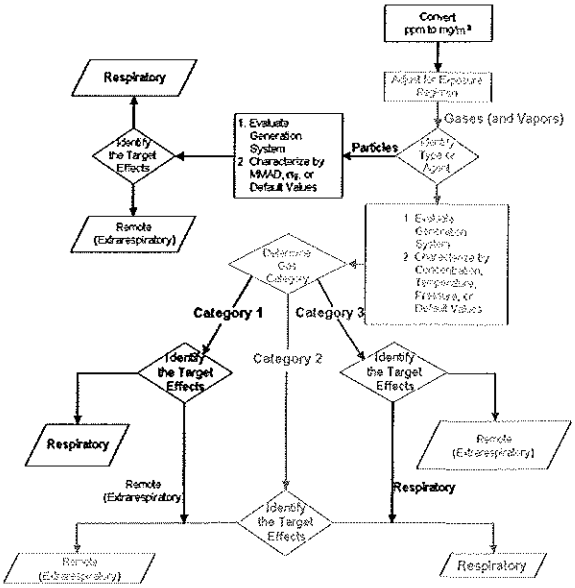
No adenocarcinomas of the nose were seen the interim groups, however; one high dose female rat that died during the course of the study had an adenocarcinoma of the nose as an incidental finding. Clearly the development of nasal tumors was only associated with very prolonged exposure to NVP. These observations are consistent with the likelihood that the tumors of the nose occurred late and only after prolonged exposure to NVP. Squamous cell carcinomas of the larynx were observed in four male and four female rats of the high exposure group after 24 months of exposure. Focal epithelial hyperplasia was seen in high exposure female and in high- and mid-exposure rats.

In summary, the inhalation of NVP vapor caused chronic toxicity and the development of neoplastic lesions in rats in all exposure groups (5-20 ppm) in an exposure-dependent manner. The most relevant non-neoplastic effects were hepatotoxicity and manifestations of local toxicity (irritancy) in the respiratory tract (nasal mucosa and larynx). The development of neoplastic lesions in the target organs; liver, nasal mucosa, and larynx is

associated with the observed chronic toxicity. The data derived from this study suggest that the prolonged exposure to NVP is required to produce a neoplastic effect.

Based on the above study, an Initial Risk Screening Level was developed from carcinogenic effects using the methodology from Rule 231. According to Rule 231 (3)(h); if exposure was by inhalation and the carcinogenic agent is a gas, the available data shall be evaluated to determine dose equivalency between humans and experimental animals. Data from the bioassay did indicate the exposure atmosphere was produced by an atomizer connected to an all-glass evaporator and heated by a thermostat-controlled water bath. The generated vapors were passed through a glass frit D2 to ensure that no aerosol reached the inhalation chambers. The new Proposed Guidelines for Carcinogen Risk Assessment (EPA/600/P-92/003C) state that the default procedure to derive a human equivalent concentration of inhaled particles and gases is by using the U.S. Environmental Protection Agency's *Method for Derivation of Inhalation Reference Concentrations and Application of Inhalation Dosimetry* guidance document (EPA/600/8-90/066F; October 1994). According to this guidance document, a key element in extrapolating laboratory animal inhalation data to humans is estimating the human equivalent concentration (HEC) or "dose" (i.e., agent mass deposited per unit surface area or tissue volume) delivered to specific target sites in the respiratory tract or made available to uptake and metabolic processes for systemic distribution. This is considered with mechanistic determinants of toxicant-target interactions and tissue responses. The HEC is the basis for comparison and choice of the critical effect and study. Calculating a HEC is a stepwise procedure. First, adjustment factors are used to determine the observed exposure effect levels in laboratory animals to estimate a concentration that would be an equivalent exposure to humans). The next step is converting the exposure regimen of the experiment to that of the human exposure scenario; that is, a continuous (24-h/day) lifetime (70-year) exposure. Then, dosimetric adjustments are appropriately applied for the type of toxicant being assessed (particle or gas, and if a gas, what category) and the effect to be assessed (respiratory tract or extra-respiratory toxicity) resulting from an inhalation exposure. Identification of the target effect(s) is also used to further define the gas category. (*Category 1 gases are defined as gases that are highly water-soluble and/or rapidly reactive in the respiratory tract. Reactivity is defined to include both the propensity for dissociation as well as the ability to serve as a substrate for metabolism in the respiratory tract. Gases in Category 1 are distinguished by the property that the gas does not significantly accumulate in the blood which would reduce the concentration driving force into the respiratory tract tissue and hence reduce the absorption rate. Gases in Category 2 are defined as gases that are moderately water-soluble that may be rapidly reversibly reactive or moderately to slowly irreversibly reactive in respiratory tract tissue. These gases are "transitional" gases that have the potential for significant accumulation in the blood and thus have the potential for both respiratory and remote (extrarespiratory) toxicity. The accumulation in the blood will reduce the concentration driving force during inspiration and thereby reduce the absorption rate or dose upon inhalation. These types of gases also have the potential for significant desorption during exhalation. Gases or vapors in Category 3 are relatively water insoluble and unreactive in the Extrathoracic and Tracheobronchial regions. Thus, the relatively limited dose to these respiratory tract regions does not*

*appear to result in any significant toxicity, although some respiratory tract toxicity may be related to recirculation. The uptake of these gases is predominately in the pulmonary region and is perfusion limited. The site of toxicity is generally remote to the principal site of absorption in the pulmonary region. (For gases in Category 3 that exhibit their toxic effects outside of the respiratory tract, an approach for the scenario when the concentration of the gas in the animals is periodic with respect to time is recommended.)* For gases, the determination of the appropriate gas category is required to determine which dosimetric adjustment would apply to calculate an HEC. A flowchart presented below, better describes the decision making process used to determine the methodology to calculate the HEC for NVP.




Utilizing the above guidance, NVP was classified in the following manner in order to determine a HEC. The first assumption was determining whether the compound is a particle or gas. Study data indicated that NVP is a gas. Next, a determination was made as to what category of gas NVP is. In reviewing the bioassay, it was determined that NVP is a Category 2 gas; or a gas that has the potential for significant accumulation in the blood and thus have the potential for both respiratory and remote (extrarespiratory) toxicity.

Study results indicated that this compound affected the nasal mucosa with an increase in nasal inflammatory lesions and nasal olfactory epithelial degeneration, in addition to causing histopathologic lesions in the liver. Therefore, this compound was classified as a Category 2 gas. However, derivations of equations regarding Category 2 gases in the 1994 Methods document are in error. The US EPA (Mark Greenberg, Hazardous Pollutant Assessment Group; National Center for Environmental Assessment) recommends a default for chemicals that would be expected to have only extrarespiratory or remote effects to use Category 3. An added assumption when considering a Category 3 gas is to determine whether the concentration of the inhaled compound within the animal achieved periodicity with respect to time (i.e., periodic steady state - the concentration versus time profile is the same for every week). It was impossible to tell

from the study the exact time course for periodicity. According to the guidance document, if the periodicity is unknown, the default value will be equal to 1 (one).

***Adjust for Exposure Regimen***

$NOAEL_{[ADJ]}$ = E ($mg/m^3$) x D (h/24h) x W (days/7 days)

E = experimental dose level
D = number of hours exposed/24 h; and
W = number of days of exposure/7 days

$Dose_{[ADJ]}$ = 0, 22.7, 45.5, 90.9 $mg/m^3$ x 6h/24h x 5days/7days

$Dose_{[ADJ]}$ = 0, 4.1, 8.1, 16.2 $mg/m^3$

***Dosimetric Adjustments and Calculation of $NOAEL_{[HEC]}$***

$$[HEC] = Dose_{[ADJ]} \times \frac{(H_{b/g})_A}{(H_{b/g})_H}$$

[HEC] = the effect level obtained with an alternative approach, dosimetrically adjusted to an HEC;

$Dose_{[ADJ]}$ = described above; and

$(H_{b/g})_A/(H_{b/g})_H$ = the ratio of the blood:gas (air) partition coefficient of the chemical for the laboratory animal species to the human value. The value of 1.0 is used for the ratio if $(H_{b/g})_A > (H_{b/g})_H$.

$$[HEC] = 0, 4.1, 8.1, 16.2\ mg/m^3 \times \frac{1}{1}$$

[HEC] = 0, 4.1, 8.1, 16.2 $mg/m^3$

The highest q1* value was produced by the data from nasal adenomas in Sprague-Dawley male rats. Doses were adjusted to inhalation doses. The number of animals per group could not be adjusted to include only those rats surviving until the time of first tumor appearance. A printout of the Global 82 model input and output is attached.

**Model Input for male rat nasal adenomas**
4,3,0,2,3,0,0
70,60,60,60
0,4.1,8.1,16.2
0,8,9,10
.01 2,.1D-4 2,.1D-5 2

MLE dose on $1x10^{-6}$ risk = 5.80E-5
95% Upper Confidence Limit on Risk = 1.35E-6

$$q_1^* = \frac{1.35E\text{-}6}{5.80E\text{-}5} = 2.3 \times 10^{-2}\ (mg/m^3/day)^{-1}$$

Utilizing the unit risk value for the nasal adenomas in male rats, an ambient air concentration corresponding to an increased cancer risk of one in a million ($1x10^{-6}$) was determined as follows:

$$\text{Concentration} = \frac{1x10^{-6}}{2.3 \times 10^{-2}\ (mg/m^3/day)^{-1}} = 0.000043\ mg/m^3$$

**Conversion of $mg/m^3$ to $ug/m^3$**

$$0.000043\ mg/m^3 \times \frac{1000\ ug}{1\ mg} = 0.04\ ug/m^3$$

**The IRSL for N-vinylpyrrolidinone-2 = 0.04 $\mu g/m^3$ based on an annual averaging;**

**The SRSL for N-vinylpyrrolidinone-2 = 0.4 ug/m3 based on an annual averaging.**

**References:**


1. Klimisch, HJ et al. 1997. Long-term inhalation toxicity of n-vinylpyrrolidinone-2 vapors. Studies in rats. Food and Chemical Toxicology, 35:1041-1060.

2. Klimisch, HJ et al. 1997. Subchronic inhalation and oral toxicity of n-vinylpyrrolidinone-2 vapors. Studies in rodents. Food and Chemical Toxicology, 35:1060-1074.

3. TSCA 8e submittal. 1992. Support: two year inhalation study with N-vinylpyrrolidinone-2 as a vapor in Sprague-dawley rats with cover letter dated 102192. Submitting organization: BASF Corp. OTS0509748-8 or 89-930000032.