Biochemical Pharmacology, Vol. 30, No. 10, pp. 1155-1157, 1981
Printed in Great Britain.

0006-2952/81/101155-07$02.00/0.
Pergamon Press Ltd.

# POTENTIATION OF HYDRALAZINE-INDUCED CONVULSION BY ISONIAZID IN RATS

Tetsuo Satoh*, Shuichi Hara, Yoko Numata and Haruo Kitagawa

Laboratory of Biochemical Pharmacology and Biotoxicology,
Faculty of Pharmaceutical Sciences, Chiba University,
Chiba, Japan

(*Received* 10 *February* 1981; *accepted* 27 *February* 1981)

Although many reports indicate side effects of hydralazine (HP) in clinical use, such as arteritis and systemic lupus erythematosus (1-3), no report on central nervous system toxicity has appeared. On the other hand, it is well established that isoniazid (INH) causes convulsion or seizure in experimental animals at high doses, which may be attributed to the decrease in gamma-aminobutyric acid (GABA) level in the brain(4,5).

The present study was designed to investigate the toxic effect of HP on the central nervous system by using convulsion and mortality as indicators in rats given HP alone and in combination with INH.

## Materials and Methods

Male Wistar rats weighing 180-200 g were used. They were housed in an air-conditioned room (23°C) and allowed free access to food and water. HP hydrochloride was purchased from Sigma Chemical Co., Inc., St. Louis, MO. HP was dissolved in saline and administered to rats intravenously or intraperitoneally. INH was purchased from Daiichi Pure Chemical Co., Tokyo, Japan, and was dissolved in distilled water. INH was orally administered at a dose of 250 mg/kg 30 min prior to HP treatment. All other chemicals were purchased from Wako Pure Chemical Co., Tokyo, Japan. GABA level in the brain was determined by the method of Löscher and Frey (4).

## Results

As can be clearly seen from Table 1, the incidences of HP-induced convulsion and death were markedly increased by pretreatment with INH. It is of interest that the abnormal behavior known as "Kangaroo posture" was observed in rats treated with HP alone and HP plus INH, whereas, INH alone did not produce any change in the general behavior of the animals at this dose level (250 mg/kg). Table 1 also shows that the doses of HP sufficient to produce convulsion and death of all the animals when administered as HP alone and as HP in combination with INH were 60 mg/kg and 40 mg/kg, respectively

Furthermore, the latent period to the onset of convulsion and the time to death were significantly shortened in the combined group, as compared with the groups given HP alone (Fig.1).

As shown in Table 2, there was no significant difference between the brain GABA levels in rats treated with HP alone and with HP in combination with INH at each time determined. However, the brain GABA level in the INH-treated group was decreased to 91.8% of the control level 30 min after pretreatment with INH, corresponding to zero time of HP treatment.

* To whom reprint requests should be addressed.

Table 1. Incidences of convulsion and death after treatment of rats with HP alone and in combination with INH

| Dose of HP (mg/kg, i.v.) | HP alone | | | HP plus INH | | |
|---|---|---|---|---|---|---|
| | S.C.C. | T.C. | Death | S.C.C. | T.C. | Death |
| 10 | 0/5 | 0/5 | 0/5 | 1/20 | 0/20 | 0/21 |
| 20 | 0/5 | 0/5 | 0/5 | 6/9 | 4/9 | 3/9 |
| 30 | 0/10 | 0/5 | 0/10 | 8/10 | 8/10 | 7/10 |
| 40 | 4/15 | 1/15 | 3/15 | 10/10 | 3/10 | 10/10 |
| 50 | 6/10 | 0/10 | 6/10 | 9/9 | 1/9 | 9/9 |
| 60 | 10/10 | 1/10 | 10/10 | 10/10 | 5/10 | 10/10 |

INH (250 mg/kg, p.o.) was administered 30 min before HP.
S.C.C.: Severe clonic convulsion; T.C.: Toxic convulsion.

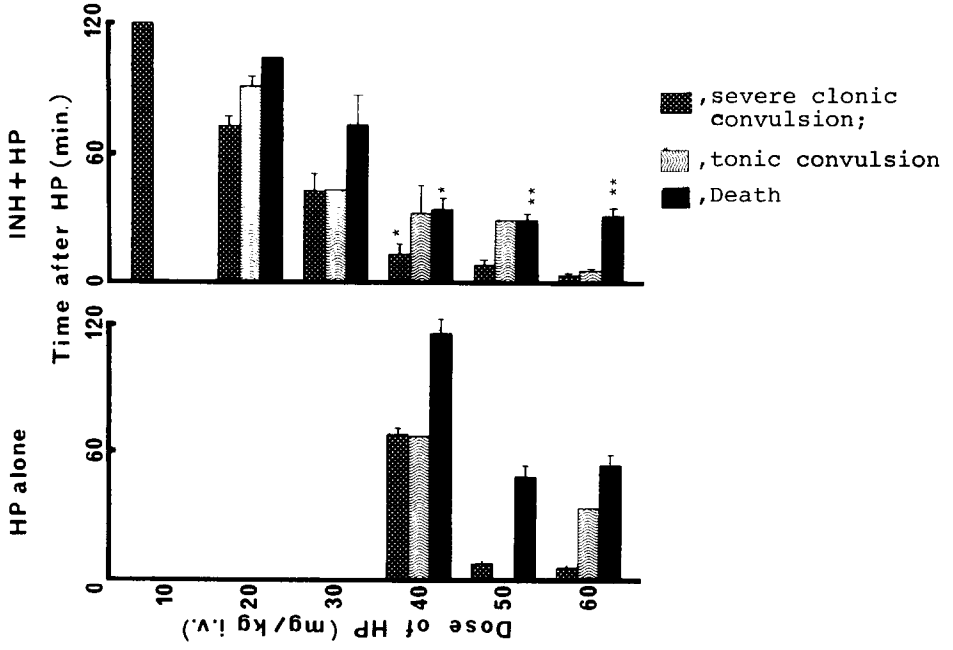


FIGURE 1. Changes in the times to convulsion and to death after i.v. treatment of rats with HP alone and in combination with INH. *P<0.001 vs HP alone, ** P<0.01 vs HP alone.

Table 2. Effect of treatment of rats with HP alone and in combination with INH on brain GABA level in rats

| Treatment | Time after HP (min) | | | |
|---|---|---|---|---|
| | 0 | 30 | 60 | 90 |
| Control | 635±7(100) | 614±23(100) | 647±14(100) | 618±19(100) |
| HP alone | --- | 518±20(84.4)* | 610±19(94.3) | 552±17(89.3) |
| HP + INH | 582±20(91.8) | 551±19(89.7) | 579±26(89.5) | 570±11(92.2) |

Figures indicate GABA μg/g brain. INH (250 mg/kg, p.o.) was administered 30 min before HP treatment (20 mg/kg, i.p. ).
*P<0.05 when compared with control. Figures in parentheses indicate percent of the control.

## Discussion

HP was known to prolong the hypnotic effect of barbiturates (6), and our previous report suggested that this effect of HP might be due to the enhancement of brain susceptibility by HP (7). In fact, rats became apparently sedated when administered HP alone at lower doses. However, as shown in this paper, HP can produce convulsion in rats at high doses or even at lower doses when combined with a non toxic dose of INH. INH has been reported to inhibit enzymatic acetylation of HP competitively in vitro (8). Table 1 and Figure 1 show that the incidences of convulsion and mortality were increased and the duration to onset of convulsion was shortened with increasing doses of HP. It is of interest that the abnormal behavior known as "Kangaroo posture" was observed only when HP alone or in combination with INH was administered to rats, and was not observed in the animals treated with INH alone at doses sufficient to induce convulsion (data not shown). Therefore, it appeared that the convulsion observed in rats after both HP and INH treatment could be attributed to the effect of HP, not INH itself, on the central nervous system.

Many earlier reports have demonstrated that hydrazine and its analogs cause convulsion when administered to experimental animals, and suggest that this effect may be related to the changes in GABA level and synthesis in the brain (9,10). Abraham and Wood also suggested that the intensity of convulsion caused by hydrazines, such as semicarbazide, increased in proportion to the decrease of GABA level in the brain (11). In the present study, it is difficult to assume that convulsion induced by HP plus INH treatment could be attributed to the change in brain GABA level (Table 2), but the decrease in GABA level at zero time after HP treatment may decrease the convulsion threshold of the rats, leading to the increase in the incidences of convulsion and death.

With regard to the occurrence of abnormal behavior of the rats, L-2,4-diaminobutyric acid, a GABA analog, is reported to produce a neurological syndrome resembling "Kangaroo posture" and convulsion (12,13).Moreover, this compound seems to act as a "false transmitter" for the neuronal uptake, storage and release of GABA in the brain (14). These findings strongly suggest that the effect of HP on the GABA system is different in nature from that of other hydrazine analogs.

Further work is in progress in this laboratory.

Acknowledgement: This work was supported in part by a Grant-in-Aid from the Ministry of Education, Science and Culture, Japan.

## References

1. H.M. Perry,Jr., A. Sakamoto and E.M. Tan, J. Lab. Clin. Med. 70, 1020 (1967).
2. H.M. Perry,Jr., E.M. Tan and S. Carmody, J. Lab. Clin. Med. 76, 114 (1970).
3. J.J. Condemi, D. Moore-Jones, J.H. Vaughan and H.M. Perry,Jr., New Engl. J. Med. 276, 286 (1967).
4. W. Löscher and -H.H. Frey, Naunyn-Schmiedeberg's Arch Pharmakol. 269, 263 (1977).
5. M. Matsumoto, M. Abe, M. Hoshino and T. Sakurai, Biochem. Pharmacol. 28, 2785 (1979).
6. J.O. Muller and J.R. Fouts, Biochem. Pharmacol. 14, 305 (1965).
7. S. Hara, T. Satoh and H. Kitagawa, Res. Commun. Chem. Pathol. Pharmacol. in press.
8. J.M. Jenne, J. Clin. Invest. 44, 1992 (1965).
9. K.F. Killam and J.A. Bain, J. Pharmacol. Exp. Ther. 119, 255 (1957).
10. K.F. Killam, J. Pharmacol. Exp. Ther. 119, 263 (1957).
11. D.E. Abraham and J.D. Wood, J. Neurochem. 17, 1197 (1970).
12. R.M. O'Neal, C. Chen, C.S. Reynolds, S.K. Meghal and R.E. Koepper, Biochem. J. 106, 699 (1968).
13. F. Vicanco, F. Ramos and C. Jimenez-Diaz, J. Neurochem. 13, 1461 (1966).
14. F. Weitsch-Dick, T.M. Jessell and J.S. Kelly, J. Neurochem. 30, 799 (1978).