Acute Toxicity of Iomex—a Petroleum Weedicide

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INTRODUCTION

Petroleum fractions containing high concentrations of toxic olefins and aromatics are well know to act as general defoliants. However, by altering the proportion of such active constituents, fractions with selective action can be obtained (DEONG, 1948). Recently a new petroleum fraction, Iomex, has been found to be an effective and economical non-specific herbicide (ANONYMOUS, 1974). Prior to its use it is obligatory to determine its toxicity. A study of acute toxicity of this preparation was, therefore, undertaken in a number of laboratory animals. The results are reported below.

MATERIALS AND METHODS

Iomex is a by-product of petroleum industry produced by M/s Indian Oil Corporation, India. It is a light pale liquid with physico-chemical properties summarized in Table 1.

Experimental Animals

The animals used for this study were: albino mice weighing 17 to 23 g; albino rates weighing 175 to 210 g; guinea pigs weighing 225 to 275 g and albino rabbits weighing 1.5 to 2.5 kg. Four animals in each group were set apart as controls and were treated in all details similarly as the experimental animals except that normal saline was used in place of Iomex.

Routes of Administration

Intraperitoneal: Iomex was injected intraperitoneally in the various animals in dose ranges shown below: mice, 5 to 15 ml; rats, 5 to 10 ml; guinea pigs, 5 to 15 ml and rabbits, 1.5 to 7 ml/kg body weight.

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Table 1

Physico-Chemical Properties of Iomex Used in This Study

Olefins Aromatics	88%
Water Content, % Vol.	Trace
Ash Content, % Wt.	0.002
Density, 15°C, gm/ml	0.8979
Carbon Residue, Ramsbottom % Wt.	0.13
b MC_1	60
Flash Point (Abel), °C	43
Gross Calorific Value, Btu/lb.	19,880
Inorganic Acidity, mg KOH/gm	Nil
Viscosity Red Wood No. 1 @ 37.8°C	28 Secs

% Distilled (IBPC Distillation)	Temp. (°C)
Initial	156
10%	181
20%	192
30%	200
40%	208
50%	218
60%	228
70%	238
80%	248
90%	260
FBP°C	290
Recovery, % Vol.	98.0
Residue, % Vol.	1.8
Loss, % Vol.	0.2

Intratracheal: This route of administration was used only in rats. Rats were anaesthetised with ether. A half inch long incision was made through the skin and subcutaneous tissue overlying the trachea immediately below the larynx. The muscles of the neck were separated in the middle line to expose the upper trachea. Iomex was injected in doses ranging from 0.25 to 1 mg/kg using 1-inch long 26-gauge needle. After injection the skin was sutured with cotton thread and animals allowed to recover.

Gastric intubation: Mice and rats were administered Iomex orally in doses ranging from 5 to 20 ml/kg with the help of 2" long blunt ended steel cannula passed down the oesophagus.

Rabbits were administered Iomex in doses ranging from 5 to 12 ml/kg. They were immobilised and the mouth was held open with a gag. The no. 4 rubber catheter was passed through a hole in the centre of the gag through the mouth down to the stomach.

The animals were observed for any signs of toxicity over a period of seven days. Motor incoordination of mice and rats was tested by the "Rota Rod" method of DURHAM and MIYA (1957), while spontaneous activity was measured by using the photocell counter of DEWS (1953). Animals dying during the period of observation were subjected to postmortem examination and any gross pathology observed was recorded.

Effect on Eye Mucosa

One-tenth ml of Iomex was instilled into the left eye of two female rabbits; the other eye served as control. In another experiment 0.1 ml of Iomex was instilled in two female rabbits daily for 5 days and the animals observed every day for a period of 15 days.

Determination of Transaminases

For serum and liver transaminase activity female albino rats were divided into four groups having 8, 4, 4 and 4 animals each. First group served as control and was injected with normal saline. Groups 2, 3, and 4 were given a single dose of 5 ml/kg of Iomex intraperitoneally and sacrificed after 3 hours, 18 hours and 5 days, respectively. Glutamic oxaloacetic (GOT) and glutamic pyruvic transaminases (GPT) in blood and liver were measured as described by REITMAN and FRANKEL (1957).

RESULTS

Table 2 summarises data with respect to per cent mortality in all the animals receiving Iomex by different routes of administration.

Mice, rats and guinea pigs tolerated fair amounts of Iomex by oral and intraperitoneal routes. There was no mortality among mice or guinea pigs at 5 ml/kg, while among rats only one out of 20 animals died at this dose level (see discussion). Among rabbits, however, there was 50 per cent mortality at this dose level by intraperitoneal route. None of the surviving animals showed any overt changes in behaviour or appearance at this dose

Table 2

Per cent Mortality among Mice, Rats, Guinea Pigs and Rabbits Given Iomex by Different Routes

Species & sex		No. of animals	Route	Dose ml/kg	Mortality %
Mouse	M	10	ip	5.0	0
		10		7.5	30
		10		10.0	50
		10		15.0	90
	F	7		5.0	0
		7		7.5	20
		8		10.0	62
		7		15.0	100
	M	6	Oral	5.0	0
		8		10.0	37
		8		15.0	87
		8		20.0	100
	F	8		5.0	0
		8		10.0	37
		8		15.0	75
		8		20.0	100
Rat	M	5	ip	5.0	0
		5		7.5	20
		5		10.0	40
	F	5		2.5	0
		5		5.0	20
		5		7.5	20
		6		10.0	33
	M	5	Oral	5.0	0
		5		10.0	40
		4		15.0	50
		5		20.0	100
	F	6		5.0	0
		5		10.0	20
		6		15.0	33
		6		20.0	82

Table 2 (contd.)

Species & sex		No. of animals	Route	Dose ml/kg	Mortality %
	F	5	i/tracheal	0.25	20
		6		0.5	33
		6		1.0	67
Guinea Pig	M	5	ip	5.0	0
		5		7.5	20
		5		10.0	40
		5		15.0	100
Rabbit	F	4	ip	1.5	0
		4		3.0	25
		4		5.0	50
		5		7.0	100
		4	Oral	5.0	0
		4		6.7	0
		4		10.0	75
		4		12.0	100

level. With higher doses (7.5 to 10 ml/kg) the most common symptoms noticed were related to CNS depression characterised by decreased spontaneous motor activity (as evidenced by "Rota Rod" and "photocell counter technique") and reduced resistance to handling. The animals, however, retained reaction to external stimuli at these dose levels.

With lethal doses, after initially decreased spontaneous activity, the most striking observation was loss of response to painful stimuli, appearing first in the hind quarters, spreading on to the forelimbs. This was followed by total loss of motility, loss of corneal reflex, deep coma and death. In animals receiving Iomex orally loose stools were occasionally observed. No rise in temperature was detected in animals given toxic doses of Iomex.

Postmortem examination of the animals receiving lethal doses of Iomex intraperitoneally revealed mild congestion of lungs, fatty degeneration of liver and hyperaemia of kidneys. Animals receiving Iomex orally showed congestion of intestines which were filled with gas. No gross abnormality was observed in any other organ.

Direct intratracheal instillation of Iomex in rats produced immediate signs of pulmonary injury characterised by dyspnea, rapid breathing, cyanosis and frothy discharge from the mouth and nostrils; the severity of symptoms were dose related. With higher doses exophthalmos was quite common at the time of death. There was mortality rate of 67% (4 out of 6) with as little as 1 ml/kg of Iomex by this route. On postmortem examination the lungs showed varying degrees of haemorrhage.

Effect on Eye Mucosa

Immediately after instillation of 0.1 ml of Iomex into the rabbit eye, congestion of the conjunctiva was observed which disappeared in a few minutes. On repeated cropping of Iomex conjunctivits developed by the 3rd day and hair around the inner canthus started falling. There was marked congestion and lacrimation up to the 8th day (3 days after stopping treatment). Thereafter the animals recovered slowly and hair started growing again.

Effect on Transaminases

Table 3 summarises the results of estimation of liver and serum transaminases of rats given 5 ml/kg of Iomex. Whereas there was no adverse change in liver GOT and GPT were significantly increased at 3 and 18 hour intervals, respectively.

DISCUSSION

In view of the nature of the material, being a petroleum fraction with IBP 156°C and FBP 290°C, exact LD_{50} of Iomex was not determined. The present data, however, suggest that LD_{50} of Iomex for mice, rats and guinea pigs by oral or intraperitoneal route will be above 10 ml/kg while for rabbits it may be about 5 ml/kg. By intratracheal route, however, the LD_{50} for rats is likely to be below 1 ml/kg. Five ml/kg had hardly any effect among mice, rats and guinea pigs. One female rat given 5 ml/kg intraperitoneally, which died on the seventh day did not show any signs typically seen in other rats given higher doses of Iomex. Moreover, no untoward signs were noted at this dose level among the surviving rats. We are, therefore, inclined to believe that this rat died from some extraneous cause.

The changes in liver transaminases (GOT & GPT) suggest that doses as high as 5 ml/kg of Iomex in rats may

Table 3. Transminases of Rats Given 5 ml/kg of Iomex i/p

Group	Time	No. of animals	GOT		GPT	
			Serum transaminase (μ mol/min/100 ml)			
I	0 hrs (Control)	7	8.01±0.40 (6.9 -9.8)		5.94±0.15 (4.0 -6.9)	
II	3 hrs	4	10.35± 0.51 (9.4 -11.8)	I vs II P≤0.01	6.88±0.99 (4.2 -8.8)	I vs II NS
III	18 hrs	4	9.07±0.71 (6.7 -9.3)	II vs III NS	9.88± 1.32 (6.6 -13.0)	I vs III P≤0.05
IV	5 days	4	8.82±0.2) (8.2 -9.5)	IV vs IV NS II vs IV NS III vs IV NS	5.80±0.22 (5.3 -6.3)	I vs IV NS II vs IV NS III vs IV P≤0.05
			Liver transaminase (μ mol/min/g tissue)			
I	0 hrs (Control)	8	14.28± 0.74 (11.0 -17.3)		28.06± 1.31 (22.7 -32.8)	
II	3 hrs	4	15.48± 0.39 (14.6 -16.4)	I vs II NS	32.1 ± 0.35 (31.5 -33.1)	I vs II NS
III	18 hrs	4	15.42± 1.17 (12.8 -18.2)	I vs III NS II vs III NS	34.05± 1.74 (29.6 -37.0)	I vs III NS II vs III NS
IV	5 days	4	14.05± 0.46 (13.1 -15.1)	I vs IV NS II vs IV NS	32.58± 1.16 (28.2 -34.1)	I vs IV NS II vs IV NS III vs IV NS

not cause any physiological disturbances. The slight rise in serum GOT and GPT observed in this study may, however, be a response to non-specific stress (PEARL et al., 1966).

By oral or intraperitoneal routes of administration Iomex appeared to cause its acute toxic effects through CNS depression. Increasing doses of Iomex caused graded CNS depression ranging from reduced motor activity to loss of response to painful stimuli, flaccid paralysis, comatose state resembling general anaesthesia and finally death. Animals dying following Iomex administration by either of these routes showed only mild degree of pathology in the lungs, liver or kidneys. On the other hand, when instilled directly into the trachea, Iomex caused extensive haemorrhages in lungs and the signs of asphyxia appeared soon after the animals recovered from anaesthesia. Therefore, it appears that two different mechanisms of Iomex are operative depending on the route of administration. Following oral or intraperitoneal administration the main mechanism appears to be the CNS depression while following intratracheal instillation interference with gaseous exchange in lungs seems to be responsible for death. This is further supported by the fact that whereas 5 ml/kg is hardly toxic to rats by intraperitoneal or oral route, 1 ml/kg is fatal to 4 out of 6 rats by the intratracheal route. The data in Table 2 provide some idea of relative toxicity of Iomex in rats by the intratracheal, oral or intraperitoneal routes. The ratio of the oral to intratracheal estimated LD_{50} of Iomex is 30:1, which is yet another instance of the well known axiom in toxicology that toxicity can be greatly modified according to the route of administration. It also indicates that greater precautions will be necessary to protect people and animals against aspiration of droplets of Iomex than against its ingestion.

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