Studies on the Improvement of Quality of Wheat Infected with Karnal Bunt. II. Nutritional and Biological Effects[1,2]

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ABSTRACT

The effect of feeding diets of wheat containing Karnal bunt and trimethylamine to rats and the effectiveness of debranning and lye peeling in controlling the adverse biological effects of the infected wheat on rats were investigated. The trials revealed that gains in body weight and protein efficiency ratio were lowest for the diet containing 10 meq of trimethylamine. Packed cell volume and neutrophils and monocytes in the blood of rats were significantly reduced by the presence of Karnal bunt or trimethylamine in the diets, while reticulocytes, leucocytes, lymphocytes, and eosinophils increased. The adverse effects of Karnal bunt on the hematological parameters were largely overcome by debranning and lye peeling. These treatments significantly reduced the activities of various enzymes in different organs and the plasma of rats from levels found for the diets containing Karnal bunt or trimethylamine.

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Karnal bunt caused by *Neovossia indica* is reported to cause discoloration of grains and to impart a fishy odor to the flour (Sekhon et al 1980b, Gopal and Sekhon 1988). The fishy odor has been attributed to the presence of trimethylamine (TMA) in the infected grains (Bedi et al 1981). Infection in wheat not only makes the products prepared from such wheat unacceptable for human consumption (Mehdi et al 1973, Sekhon et al 1980a) but also is reported to have adverse biological effects (El-Behadli et al 1978, Sharma et al 1982, Gopal et al 1988). In earlier studies (Sekhon et al 1992) we described treatments by which the adverse effects of the infected wheat on the quality and acceptability of baked products could be controlled. In the present study we report on the effectiveness of these treatments in overcoming the adverse biological effects.

MATERIALS AND METHODS

Bulk lots of wheat infected with Karnal bunt (5 and 10% bunted wheat) were prepared and treated with debranning and lye peeling, as described in the companion article (Sekhon et al 1992).

The experiment was carried out by feeding rats diets prepared from 5 and 10% untreated infected wheat and diets prepared from debranned and lye-peeled infected wheat. The sound wheat diets and those containing 5 and 10 meq of pure TMA were also evaluated. Casein was the protein source in the reference diet. The protein content of various diets was maintained at the 7% level, and fiber and crude fat was maintained at constant levels (5 and 10%, respectively). All diets were adequately supplied with minerals (Oser 1965) and vitamins (Chapman et al 1959). The composition of undebranned, debranned, and lye-peeled wheat having 0, 5, and 10% Karnal-bunt-infected grains used in different diets was described in the companion article (Sekhon et al 1992). The composition of ingredients used in different diets is given in Table I. Albino rats 24±1 days old were individually weighed and randomly distributed into 12 groups (four of each sex), each averaging similar body weights. The animals were housed individually in aluminium cages with wire mesh bottoms. Diets were given ad libitum. The animals were weighed at weekly intervals for four weeks, and protein efficiency ratio (PER) was calculated according to the procedure of Chapman et al (1959). Rats were fed continuously for two months, then sacrificed. Blood was drawn from the aorta in tubes containing heparin for determination of hemoglobin, packed cell volume (PCV), red blood cell count (RBC), white blood cell count (WBC), and differential leucocyte count (DLC). Various organs such as the liver, heart, kidney, testes, and spleen were removed, cleaned of connective tissues, and frozen for later use. Homogenate (25%, w/v) of each tissue sample was prepared in chilled distilled water. Homogenates were centrifuged and the supernatants used for enzyme analysis. Phosphatases, transferases, and lactate dehydrogenase were analyzed by spectrophotometric methods (Wootton 1964).

The data were statistically analyzed using the paired *t* test (Steel and Torrie 1960).

RESULTS AND DISCUSSION

Protein Quality

In comparison with a reference diet, feed intake was greater in rats fed diets prepared from 5 and 10% infected wheat and on diets having 5 meq of TMA. Feed intake in rats fed diets prepared from debranned and lye-peeled infected wheat did not differ significantly from the rate observed in rats fed the sound wheat diet (Table II).

The gain in body weight of rats fed different wheat diets did not differ significantly, although they differed significantly from the reference diet groups. Rats fed diets having 10 meq of TMA had significantly lower gains in body weight with respect to all wheat diets. The PER of the casein diet was significantly higher compared with other diets. The PER values of different wheat diets did not differ significantly among themselves, but the PER of the diet containing 10 meq of TMA was significantly lower.

The results indicate that the presence of infected wheat or the addition of TMA in the diets did not affect the acceptability of the diet by the rats; in fact, the feed intake was higher in the case of diets containing the diseased wheat. The results corroborate the earlier findings of Gopal et al (1988). It seems that the higher feed intake of the infected wheat diets to a considerable extent overcomes the adverse effect of the infected wheat in the gain in body weight and PER in rats because of the concomitant higher intake of nutrients. This finding is supported by the results of Gopal et al (1988), who actually found higher PER values for such diets. The lower weight gain and reduced PER values in rats fed the diet containing 10 meq of TMA point to a possible toxic effect of TMA at this concentration. Such a possibility is supported by similar earlier reports (Sharma et al 1982, Gopal et al 1988).

Hematological Studies

The hemoglobin content of rats fed diets containing 0, 5, and 10% diseased kernels treated with various treatments, as well as untreated kernels, did not differ significantly. However, a significant reduction (24.2%) in hemoglobin content was observed in rats fed diets containing 10 meq of TMA, compared to the groups fed sound wheat (Table III). PCV, neutrophiles, and

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TABLE I
Composition of Ingredients Used in Different Experimental Diets

Experimental Diets	Ingredients (g/100 g of diet)							Trimethylamine (meq/100 g of wheat)
	Casein	Wheat	Salt Mixture	Vitamin Mixture	Refined Grount-Nut Oil	Cellulose	Corn Starch	
Reference	7.0	...	4.0	1.0	10.0	5.00	73.00	...
Uninfected wheat	...	74.79	4.0	1.0	8.73	2.83	8.65	...
Uninfected wheat, debranned	...	77.00	4.0	1.0	8.72	3.57	5.71	...
Uninfected wheat, lye peeled	...	79.54	4.0	1.0	8.66	3.31	3.49	...
5% Infected wheat	...	73.76	4.0	1.0	8.67	2.76	9.81	...
5% Infected wheat, debranned	...	76.60	4.0	1.0	8.68	3.50	6.22	...
5% Infected wheat, lye peeled	...	78.92	4.0	1.0	8.63	3.27	4.18	...
10% Infected wheat	...	72.92	4.0	1.0	8.63	2.60	10.85	...
10% Infected wheat, debranned	...	76.25	4.0	1.0	8.67	3.50	6.85	...
10% Infected wheat, lye peeled	...	78.20	4.0	1.0	8.65	3.20	4.95	...
Uninfected wheat	...	74.79	4.0	1.0	8.73	2.83	8.65	5
Uninfected wheat	...	74.79	4.0	1.0	8.73	2.83	8.65	10

TABLE II
Parameters of Different Diets Fed to Rats for Four Weeks (Mean ± SE)[a,b]

	Type of Diet											
		0% Infected Wheat			5% Infected Wheat			10% Infected Wheat			Wheat with Trimethylamine	
	Reference Diet	Untreated	Debranned	Lye Peeled	Untreated	Debranned	Lye Peeled	Untreated	Debranned	Lye Peeled	5 meq/100 g of Wheat	10 meq/100 g of Wheat
Feed intake, g	97.85 a ± 4.46	122.00 b ± 4.88	120.42 b ± 5.12	115.43 b ± 2.16	139.62 c ± 5.74	118.57 b ± 5.05	126.62 b ± 4.49	130.75 bc ± 5.93	119.25 b ± 6.65	129.75 b ± 3.38	129.62 b ± 6.87	128.00 b ± 5.85
Protein consumed, g	6.85 a ± 0.31	8.45 b ± 0.34	8.43 b ± 0.35	8.08 b ± 0.15	9.77 c ± 0.40	8.30 b ± 0.35	8.86 b ± 0.35	9.15 bc ± 0.41	8.38 b ± 0.47	9.10 b ± 0.24	9.06 bc ± 0.40	8.96 bc ± 0.46
Gain in body weight, g	21.00 a ± 1.80	14.94 b ± 1.20	14.00 b ± 0.73	13.00 b ± 1.80	15.10 b ± 2.18	13.71 b ± 1.73	14.00 b ± 2.43	14.76 b ± 2.70	14.07 b ± 2.61	14.12 b ± 2.11	15.40 b ± 3.19	8.16 c ± 2.40
Protein efficiency ratio	3.06 a ±0.33	1.76 b ± 0.24	1.67 b ± 0.07	1.60 b ± 0.17	1.53 b ± 0.21	1.66 b ± 0.21	1.56 b ± 0.22	1.55 b ± 0.24	1.63 b ± 0.30	1.55 b ± 0.21	1.70 b ± 0.32	0.90 c ± 0.28

[a] Values with similar letters do not differ significantly ($P < 0.05$).
[b] Higher ranked letters are significantly different from lower ranked letters in the following order: c > b > a.

TABLE III
Parameters of Blood of Rats Fed on Sound and Bunted Wheat Pretreated with Various Treatments (Mean ± SE, $n = 4$–7)[a,b]

	Type of Diet											
		0% Infected Wheat			5% Infected Wheat			10% Infected Wheat			Wheat with Trimethylamine	
	Reference Diet	Untreated	Debranned	Lye Peeled	Untreated	Debranned	Lye Peeled	Untreated	Debranned	Lye Peeled	5 meq/100 g of Wheat	10 meq/100 g of Wheat
Hemoglobin, g/100 ml	11.70 a ± 0.97	11.10 a ±1.12	12.50 a ±1.25	12.90 a ±0.97	11.10 a ±2.12	11.60 a ±1.62	11.30 a ±0.84	11.90 a ±1.63	11.40 a ±0.75	11.30 a ±0.63	9.31 b ±0.11	8.42 c ±0.22
Packed cell volume, %	40.3 a ±1.31	39.10 a ±0.97	39.2 a ±1.69	39.5 a ±2.15	32.2 b ±1.38	38.50 a ±1.54	38.50 a ±1.99	30.40 bc ±2.02	36.10 a ±1.45	36.80 a ±1.32	28.10 c ±2.05	23.80 d ±2.62
Reticulocytes (count × $10^4/\mu l$)	612 a ±4.25	617 a ±13.71	621 a ±8.61	623 a ±9.2	693 b ±1.53	638 a ±4.85	630 a ±3.70	772 c ±3.50	645 a ±3.50	650 a ±1.73	932 d ±1.73	985 e ±1.52
Leucocytes (count/μl)	9,434 a ±74.7	9,439 a ±97.4	9,437 a ±122.2	9,364 a ±106.1	9,403 a ±87.5	9,379 a ±86.2	9,387 a ±83.1	9,949 b ±75.9	9,550 a ±76.6	9,480 a ±63.2	10,032 b ±54.1	10,718 c ±106.5
Neutrophiles	43.5 a ±2.50	45.5 a ±3.00	46.5 a ±3.50	42.0 a ±2.10	31.50 b ±1.75	36.75 a ±2.50	36.80 a ±3.50	28.50 b ±2.75	35.00 a ±2.00	35.75 a ±2.50	18.00 c ±2.00	17.00 c ±1.75
Lymphocytes	50.00 a ±3.75	50.00 a ±3.50	49.00 a ±3.00	50.50 a ±3.50	60.50 a ±3.75	56.00 a ±3.25	56.80 a ±4.00	68.50 b ±4.50	58.00 a ±4.75	57.20 a ±4.50	71.50 b ±4.50	74.50 b ±4.50
Eosinophiles	2.75 a ±0.25	2.75 a ±0.05	2.50 a ±0.05	2.50 a ±0.05	4.00 b ±0.05	3.00 a ±0.25	3.00 a ±0.25	5.75 c ±0.25	4.00 b ±0.25	3.75 a ±0.25	5.00 c ±0.25	6.25 d ±0.25
Monocytes	4.50 a ±0.25	4.50 a ±0.25	4.50 a ±0.25	4.50 a ±0.25	3.75 a ±0.25	4.00 a ±0.25	4.00 a ±0.25	2.50 b ±0.25	3.75 a ±0.25	3.75 b ±0.50	2.50 b ±0.55	1.50 b ±0.55
Basophiles	A	A	A	A	A	A	A	A	A	A	A	1 ± 0

[a] Values with similar letters do not differ significantly ($P < 0.05$).
[b] Higher ranked letters are significantly different from lower ranked letters in the following order: d > c > b > a.

monocytes were significantly reduced, whereas reticulocytes, leucocytes, lymphocytes, and eosinophiles were significantly increased by the presence of infected wheat or TMA, and the alteration was of higher magnitude in the case of diets having TMA. The alteration in monocytes, leucocytes, and lymphocytes with diets having 5% infected wheat was not significant with respect to the control wheat diet. Debranning and lye peeling of infected wheat brought the altered hematological parameters

TABLE IV
Alkaline Phosphatase Activity[a] in Various Organs of Rats Fed on Sound and Bunted Wheat Pretreated with Various Treatments (Mean ± SE, $n = 4$–7)[b,c]

	Type of Diet											
		0% Infected Wheat			5% Infected Wheat			10% Infected Wheat			Wheat with Trimethylamine	
Organ	Reference Diet	Untreated	Debranned	Lye Peeled	Untreated	Debranned	Lye Peeled	Untreated	Debranned	Lye Peeled	5 meq/100 g of Wheat	10 meq/100 g of Wheat
Liver	28.50 a ±0.37	37.40 b ±1.01	32.60 b ±0.33	30.50 b ±0.33	63.00 c ±0.47	40.20 b ±0.64	39.10 b ±1.52	105.70 d ±1.97	70.50 c ±2.07	77.50 c ±2.15	222.20 e ±3.11	331.60 e ±4.15
Heart	103.50 a ±1.52	107.20 a ±1.59	111.70 a ±1.03	109.80 a ±2.11	163.60 c ±1.33	114.40 a ±0.29	113.50 a ±0.34	209.30 d ±1.12	147.20 b ±1.17	140.10 b ±1.54	265.50 e ±1.59	415.20 f ±1.65
Lungs	34.00 a ±0.93	37.20 b ±0.32	39.50 b ±0.11	32.10 a ±0.64	44.20 c ±0.78	37.20 b ±1.04	30.50 a ±1.11	53.90 c ±1.64	38.20 b ±1.60	36.80 b ±2.15	76.70 d ±3.10	83.70 d ±3.42
Brain	93.50 a ±2.52	106.40 a ±1.34	111.10 b ±2.17	107.60 b ±2.22	165.30 d ±1.63	112.00 b ±2.13	114.00 b ±2.03	206.00 e ±2.70	112.10 b ±1.65	134.50 c ±1.77	311.30 f ±0.97	370.90 g ±3.97
Kidney	120.20 a ±3.53	137.90 a ±3.61	124.80 a ±2.11	132.70 a ±3.15	179.30 b ±2.63	143.50 a ±2.27	140.40 a ±1.63	271.70 c ±0.79	143.00 a ±2.32	176.60 b ±1.64	317.40 d ±1.27	522.90 e ±1.17
Spleen	43.10 a ±1.83	49.20 a ±1.25	45.40 a ±0.69	44.00 a ±2.11	59.10 b ±2.36	50.40 a ±2.05	48.30 a ±1.32	73.40 c ±1.51	60.00 b ±1.68	60.20 b ±1.71	88.30 c ±2.99	106.50 d ±3.17
Intestine	52.90 a ±2.15	53.70 a ±2.66	50.10 a ±2.52	55.30 a ±2.93	56.20 a ±2.11	59.50 a ±2.63	61.10 a ±2.51	59.20 a ±2.33	57.10 a ±2.17	55.20 a ±2.17	93.60 b ±2.10	95.70 b ±3.15
Plasma	53.90 a ±1.37	56.60 a ±1.26	59.30 a ±1.08	55.20 a ±1.62	77.10 b ±1.31	62.50 a ±1.35	61.30 a ±1.45	89.20 c ±1.42	73.90 b ±1.29	70.70 b ±1.53	112.10 d ±1.92	164.50 e ±2.54

[a] Nanometers of phenol liberated per minute per gram or milligram of sample.
[b] Values with similar letters do not differ significantly ($P < 0.05$).
[c] Higher ranked letters are significantly different from lower ranked letters in the following order: g > f > e > d > c > b > a.

TABLE V
Acid Phosphatase Activity[a] in Various Organs of Rats Fed on Sound and Bunted Wheat Pretreated with Various Treatments (Mean ± SE, $n = 4$–7)[b,c]

	Type of Diet											
		0% Infected Wheat			5% Infected Wheat			10% Infected Wheat			Wheat with Trimethylamine	
Organ	Reference Diet	Untreated	Debranned	Lye Peeled	Untreated	Debranned	Lye Peeled	Untreated	Debranned	Lye Peeled	5 meq/100 g of Wheat	10 meq/100 g of Wheat
Liver	53.90 a ±2.50	59.50 a ±2.56	56.20 a ±2.55	53.00 a ±2.56	67.10 b ±1.64	60.20 a ±1.36	58.50 a ±1.76	95.90 c ±1.78	62.50 a ±1.73	60.30 a ±1.78	109.20 d ±1.73	175.10 e ±1.78
Heart	13.70 a ±2.33	17.50 a ±2.15	15.30 a ±3.13	14.10 a ±3.75	21.20 b ±3.86	17.10 a ±3.11	15.90 a ±3.75	38.10 c ±4.12	25.20 b ±4.16	22.10 b ±4.12	47.40 d ±3.10	59.50 e ±3.15
Lungs	50.90 a ±2.93	59.20 a ±3.62	60.10 a ±3.92	52.50 a ±3.85	65.90 b ±2.94	55.40 a ±1.68	52.40 a ±2.68	73.20 b ±1.47	62.20 a ±2.81	59.20 a ±3.15	85.30 c ±3.11	103.70 d ±2.17
Brain	6.52 a ±2.52	6.30 a ±1.88	6.72 a ±1.63	6.47 a ±0.98	8.36 a ±2.54	7.86 a ±2.63	7.10 a ±2.68	9.35 a ±2.54	8.42 a ±3.18	8.21 a ±4.02	10.10 a ±3.50	12.55 a ±3.50
Kidney	83.50 a ±3.76	94.10 a ±2.50	95.20 a ±1.93	90.30 a ±1.85	141.50 c ±1.74	120.40 c ±1.43	110.40 b ±1.82	210.90 c ±1.76	152.20 d ±2.50	140.60 c ±3.37	322.50 f ±3.30	465.30 g ±3.65
Spleen	13.50 a ±3.54	17.30 a ±2.93	15.00 a ±3.63	15.10 ab ±2.91	22.00 ab ±4.15	17.90 a ±3.93	15.70 a ±3.16	38.40 cd ±4.12	28.20 b ±4.16	28.00 b ±4.12	47.40 b ±3.13	59.50 d ±3.15
Intestine	20.30 a ±2.13	31.40 ab ±3.75	29.50 ab ±3.65	22.80 ab ±6.53	37.30 b ±5.36	31.10 ab ±4.68	25.00 ab ±5.50	41.70 b ±3.18	38.00 b ±2.24	37.60 b ±1.17	63.70 c ±2.15	79.80 d ±2.63
Plasma	2.93 a ±2.11	3.16 a ±1.93	3.05 a ±1.93	3.19 a ±2.04	6.52 ab ±2.07	5.70 ab ±3.10	4.83 ab ±3.17	9.35 ab ±3.09	8.71 ab ±1.67	7.92 ab ±1.74	10.10 ab ±1.63	11.51 b ±1.54

[a] Nanometers of phenol liberated per minute per gram or milligram of sample.
[b] Values with similar letters do not differ significantly ($P < 0.05$).
[c] Higher ranked letters are significantly different from lower ranked letters in the following order: g > f > e > d > c > b > a.

to a level that was not significantly different from that of sound wheat.

The results clearly show that the presence of infected wheat or TMA in the diet adversely affected the hematological parameters; the effect was severe in case of the TMA diet. Debranning and lye peeling were quite effective in overcoming these effects, and the values of the various parameters after these treatments were quite comparable to those in rats fed the sound wheat diet.

TABLE VI
Lactate Dehydrogenase Activity[a] in Various Organs of Rats Fed on Sound and Bunted Wheat Pretreated with Various Treatments (Mean ± SE, $n = 4$-7)[b,c]

	Type of Diet											
		0% Infected Wheat			5% Infected Wheat			10% Infected Wheat			Wheat with Trimethylamine	
Organ	Reference Diet	Untreated	Debranned	Lye Peeled	Untreated	Debranned	Lye Peeled	Untreated	Debranned	Lye Peeled	5 meq/100 g of Wheat	10 meq/100 g of Wheat
Liver	20.20 a ±0.65	27.10 b ±0.73	22.70 a ±0.82	25.00 b ±1.11	39.40 c ±0.75	28.60 b ±0.75	27.60 b ±0.73	53.80 d ±0.18	40.00 c ±1.62	40.20 c ±1.15	63.00 e ±2.12	84.50 f ±2.15
Heart	93.10 a ±3.33	95.30 a ±2.17	97.90 a ±1.64	95.30 a ±0.63	128.80 b ±0.68	98.90 a ±1.21	98.30 a ±1.34	206.30 e ±1.92	130.00 b ±1.65	100.00 a ±1.63	322.20 f ±1.37	446.10 g ±1.75
Lungs	57.50 a ±2.11	55.50 a ±2.15	59.30 a ±3.12	56.90 a ±1.64	89.30 c ±1.29	62.70 a ±1.45	60.90 a ±1.46	103.30 d ±1.37	72.40 b ±2.11	70.50 b ±2.50	212.20 e ±2.70	299.10 f ±2.15
Brain	24.60 a ±3.15	25.20 a ±3.30	22.50 a ±3.81	21.90 a ±2.15	39.30 b ±2.25	32.30 a ±2.25	28.60 a ±1.63	57.30 c ±2.11	34.00 b ±2.30	30.10 a ±3.15	93.20 d ±2.31	103.30 e ±2.15
Kidney	40.20 a ±3.60	40.10 a ±3.52	42.20 a ±2.63	43.50 a ±2.11	59.30 b ±2.25	50.30 a ±2.45	46.90 a ±2.33	78.50 c ±2.18	62.30 b ±2.32	59.50 b ±2.54	132.30 d ±2.63	197.10 e ±2.18
Spleen	12.90 a ±3.11	13.50 a ±3.54	15.30 a ±3.63	14.40 a ±2.18	18.50 ab ±2.13	16.30 a ±3.17	15.20 a ±3.09	22.10 b ±3.09	19.50 ab ±2.11	17.30 a ±3.15	45.20 c ±3.03	57.10 d ±4.11
Intestine	37.50 a ±2.54	37.00 a ±3.59	41.50 a ±4.06	39.30 a ±3.29	53.20 b ±2.83	45.10 a ±2.99	40.50 a ±3.83	69.30 c ±2.92	53.20 b ±1.93	46.10 a ±2.95	98.50 d ±1.63	135.00 e ±1.75
Plasma	112.30 a ±1.63	112.70 a ±2.23	119.00 a ±1.15	113.40 a ±1.63	187.50 c ±1.19	120.30 a ±2.80	126.20 a ±2.72	212.10 d ±3.50	163.70 b ±2.17	128.70 a ±3.30	341.90 e ±2.15	422.50 f ±2.01

[a] Nanometers of phenol liberated per minute per gram or milligram of sample.
[b] Values with similar letters do not differ significantly ($P < 0.05$).
[c] Higher ranked letters are significantly different from lower ranked letters in the following order: g > f > e > d > c > b > a.

TABLE VII
Aspartate Amino Transferse Activity[a] in Various Organs of Rats Fed on Sound and Bunted Wheat Pretreated with Various Treatments (Mean ± SE, $n = 4$-7)[b,c]

	Type of Diet											
		0% Infected Wheat			5% Infected Wheat			10% Infected Wheat			Wheat with Trimethylamine	
Organ	Reference Diet	Untreated	Debranned	Lye Peeled	Untreated	Debranned	Lye Peeled	Untreated	Debranned	Lye Peeled	5 meq/100 g of Wheat	10 meq/100 g of Wheat
Liver	194.70 a ±3.64	187.30 a ±3.83	196.20 a ±3.45	204.10 a ±3.92	282.30 b ±3.65	206.50 a ±4.11	202.90 a ±5.32	463.10 e ±5.11	407.60 d ±3.62	300.20 c ±1.58	632.50 f ±2.59	817.40 g ±2.65
Heart	86.60 a ±1.66	88.70 a ±1.32	93.50 a ±1.06	101.10 a ±1.94	153.20 b ±1.38	94.20 a ±1.54	101.50 a ±1.32	282.00 e ±2.02	232.10 d ±2.11	193.40 c ±3.15	326.50 f ±3.66	439.30 g ±2.54
Lungs	119.30 a ±2.16	136.20 b ±2.11	141.10 b ±1.44	132.40 b ±1.38	193.50 c ±1.54	114.20 b ±1.93	144.10 b ±1.64	271.70 d ±2.32	198.00 c ±2.15	158.60 b ±2.02	327.80 e ±1.98	439.90 f ±1.63
Brain	231.20 a ±4.57	249.10 b ±3.92	244.90 b ±2.53	264.70 b ±4.11	340.40 c ±3.25	270.20 b ±4.10	260.20 b ±4.50	490.10 d ±3.11	334.50 c ±2.06	321.30 c ±2.73	811.20 e ±2.56	933.10 f ±3.64
Kidney	133.40 a ±3.11	137.10 a ±2.44	132.70 a ±4.31	139.60 a ±2.43	260.20 d ±2.16	140.10 a ±2.25	140.50 a ±3.16	380.20 f ±2.32	217.90 c ±1.99	185.80 b ±1.93	411.70 e ±2.10	538.60 f ±3.55
Spleen	88.20 a ±1.93	94.10 ab ±1.34	88.60 a ±2.15	87.20 a ±2.62	186.90 c ±1.93	100.30 b ±2.65	90.30 b ±2.10	283.10 e ±3.01	230.40 d ±4.15	180.60 c ±4.25	445.50 f ±5.13	511.00 g ±5.55
Intestine	53.50 a ±2.13	59.30 ab ±1.27	52.10 a ±2.36	57.80 a ±3.54	79.90 c ±3.74	60.00 b ±3.71	59.50 ab ±3.75	160.20 d ±3.10	82.10 c ±2.06	80.80 c ±2.15	216.90 e ±2.63	323.20 f ±2.25
Plasma	86.30 a ±3.45	87.50 a ±3.46	88.40 a ±4.13	93.60 a ±4.15	178.70 c ±5.10	132.10 b ±6.11	104.90 a ±5.40	273.00 e ±5.36	222.60 d ±5.25	164.50 c ±3.12	327.00 f ±2.22	388.30 g ±2.74

[a] Nanometers of phenol liberated per minute per gram or milligram of sample.
[b] Values with similar letters do not differ significantly ($P < 0.05$).
[c] Higher ranked letters are significantly different from lower ranked letters in the following order: g > f > e > d > c > b > a.

TABLE VIII
Alanine Amino Transferase Activity[a] in Various Organs of Rats Fed on Sound and Bunted Wheat Pretreated with Various Treatments (Mean ± SE, $n = 4$–7)[b,c]

Organ	Type of Diet											
		0% Infected Wheat			5% Infected Wheat			10% Infected Wheat			Wheat with Trimethylamine	
	Reference Diet	Untreated	Debranned	Lye Peeled	Untreated	Debranned	Lye Peeled	Untreated	Debranned	Lye Peeled	5 meq/100 g of Wheat	10 meq/100 g of Wheat
Liver	345.30 a ±5.93	350.10 a ±4.95	350.30 a ±6.14	351.90 a ±5.96	475.40 b ±7.79	354.60 a ±8.51	352.40 a ±9.61	593.00 d ±7.32	510.10 c ±2.15	460.70 b ±3.18	871.40 e ±2.94	1022.20 f ±2.73
Heart	39.20 a ±1.12	42.50 a ±2.19	42.30 a ±3.11	47.50 b ±1.05	69.60 c ±2.12	50.20 b ±2.15	48.10 b ±3.30	103.70 d ±3.18	79.50 c ±3.22	67.30 c ±3.62	229.00 e ±2.35	311.50 f ±2.15
Lungs	95.50 a ±3.92	103.90 a ±2.96	107.40 a ±2.68	91.60 a ±2.53	183.40 d ±1.65	106.00 a ±3.52	110.10 a ±2.58	219.90 e ±2.93	165.20 c ±2.58	134.30 b ±5.53	447.50 f ±5.23	577.40 g ±4.32
Brain	43.30 a ±0.40	47.70 a ±0.44	45.90 a ±0.35	50.30 b ±0.69	73.10 d ±0.29	60.00 c ±0.36	50.60 b ±0.21	93.00 e ±0.29	80.30 d ±0.52	62.40 c ±0.18	162.60 f ±0.31	227.50 g ±0.34
Kidney	126.20 a ±2.55	137.30 ab ±3.62	138.50 ab ±3.98	128.10 a ±2.55	199.20 c ±1.99	146.30 b ±2.36	132.10 ab ±2.84	260.20 d ±2.74	180.20 c ±1.65	142.20 b ±3.12	387.40 f ±5.93	596.10 g ±5.63
Spleen	63.20 a ±3.63	60.10 a ±3.45	65.00 a ±2.11	72.10 a ±2.52	98.70 b ±0.42	70.00 a ±1.80	70.30 a ±1.52	157.20 c ±2.27	100.10 b ±2.80	93.00 b ±2.68	370.60 d ±2.53	485.40 e ±2.63
Intestine	18.60 a ±0.37	17.80 a ±0.48	19.90 a ±0.53	20.10 a ±0.41	63.60 b ±0.45	22.40 a ±0.63	20.50 a ±0.42	96.20 c ±0.45	60.10 b ±0.41	60.10 b ±0.43	209.70 d ±0.39	324.30 e ±0.42
Plasma	40.10 a ±0.52	42.50 a ±1.32	39.30 a ±1.62	45.20 a ±2.15	83.10 b ±2.66	42.90 a ±3.33	40.50 a ±4.63	116.20 c ±4.82	90.30 b ±5.12	82.30 b ±6.25	234.10 d ±6.33	357.00 e ±7.18

[a] Nanometers of phenol liberated per minute per gram or milligram of sample.
[b] Values with similar letters do not differ significantly ($P < 0.05$).
[c] Higher ranked letters are significantly different from lower ranked letters in the following order: g > f > e > d > c > b > a.

Enzyme Activity

Alkaline phosphatase activity in various organs (except the intestine) of rats fed diets containing diseased wheat was significantly higher than in those fed sound wheat diets (Table IV). The activity decreased significantly when diseased wheat was used after debranning or lye peeling. The presence of TMA in the diet appreciably increased the activity of alkaline phosphatase in all organs. An increase of 8.9, 3.9, 2.3, 3.6, and 2.2 times the activity by 10 meq of the TMA diet was observed in the liver, heart, lung, brain, kidney, and spleen, respectively, compared to similar tissues from rats fed sound wheat. Acid phosphatase activity (Table V) also showed a similar pattern as that of alkaline phosphatase with respect to use of diseased wheat, treated wheat, and the addition of TMA to the diet. The increase in acid phosphatase activity in intestine and plasma was significant only in the case of diets containing TMA in comparison to the sound wheat diet.

Various organs (except the spleen) of rats fed diets containing Karnal-bunt-infected wheat also had significantly higher lactate dehydrogenase activity than did those of rats fed sound wheat diets (Table VI). Diseased grains in the diet also increased the lactate dehydrogenase activity of plasma. Addition of 5 and 10 meq of TMA in diets further elevated the lactate dehydrogenase activity in various organs.

The results indicate that the activity of various enzymes (Tables VII and VIII), including transferases in tissues and plasma, increased with the feeding of Karnal-bunt-infected grain in comparison to that for control samples. The activities were even higher in the case of diets containing pure TMA. The activities were considerably reduced by debranning or lye peeling, and the reduction in most of cases was to the extent that the differences in levels with respect to controls became nonsignificant. The increased activity of enzymes in diets containing infected grains might be due to adverse effects of the fungal mass, TMA, and/or phenolic compounds on the organs. The elevation of enzyme activity in the blood may be due to leakage of these elevated enzymes from various tissues to the blood stream (Rouiller 1964). However, the much higher enzyme activity in TMA-containing diets suggests that TMA might be the primary cause of the increased activity in tissues and plasma. Free TMA seems to be more effective in this respect. Higher activities of enzymes in tissues and plasma under the influence of various biological toxicants have been reported (Abderhalden 1969, Zimmerman and Henry 1969, Kaplan and Righethi 1970).

CONCLUSION

Feeding rats diets containing wheat infected by Karnal bunt results in increased activity of enzymes in various tissues and affects a number of hematological parameters. TMA may be one of the major causes of the increased activity in tissues and plasma. Debranning and lye peeling are quite effective in overcoming these adverse effects.

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