clearly argues against a possibility of restrictive entrance of the oxidized stain inside the relatively dense, microvesicular substructure of the apical corpuscle. Significantly, in the initial phase of the experiments, the whole tip of vegetative hyphae first takes on a bluish tint showing, as above with the pH indicators, that the stains can freely penetrate inside the spherical organelle, in which they either switch in their colour for pH reasons or, as with neutral red or methylene blue and additionally with Janus green B (white to pinkish 'Spitzenkörper' contrasting with the densely green mitochondria), as a consequence of their local reduction. To further ascertain whether the 'Spitzenkörper' is well differentiated in its permeability characteristics from the lipid granules, which can occasionally migrate close to the hyphal tips, especially in those of *Monilia*[9], vegetative hyphae were counterstained in Sudan III. Conclusively, in all actively elongating hyphae, a discrete white corpuscle could be seen in the ultimate tips, in clear contrast with the orange stained subapical lipid granules. Only in vacuolated hyphae, somewhat arrested in their growth, one stained granule could occasionally reach the tip.

A final question is what sustains the reducing power among the microvesicula of the 'Spitzenkörper'? It might be either NADH overproduced by apical glycolysis, further excreted as reducing equivalents of ethanol by vegetative hyphae at least in *Neurospora*[13] and/or -SH groups associated with the enzymatic proteins of this alcohol glycolysis[7]. In favour of the local accumulation of both types of these reducing compounds, we can make the following preliminary observations: a) a positive blue reduction of a direct acceptor of the NADH electrons[12], phenazine methosulfate (PMS), visibly expressed by a generalized greenish blue tinge in the apices of *Neurospora* hyphae ('Spitzenkörper' dispersed by the lethal, 10^{-4}, concentration of the yellow water solution of PMS) while a faint but more frankly blue reaction had been obtained with the *Allomyces* hyphal tips[7]; b) a positive reaction of Na nitroprusside revealed by a few dark reddish-violet deposits on the central part of apices elongating from wide hyphae of *Monilia* bathed in a 10^{-4} solution of this classical[14] and normally not too sensitive reagent for -SH groups.

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RNA polymerisation capacity and permeability to ribonucleoside triphosphates of nuclei from livers of whole-body X-irradiated rats

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Summary. For 4–18 h following whole-body X-irradiation of rats, liver nuclei showed a progressive increase in the permeability to ribonucleoside triphosphates (as assessed in vitro using tritiated uridine triphosphate (UTP)) and in the capacity to polymerise RNA in vitro (Mg^{++}-containing and $Mn^{++}/(NH_4)_2SO_4$-containing assay systems).

Whole-body radiation exposure of animals has been shown to stimulate the rate of RNA synthesis in the liver[1-3]. The stimulus in RNA synthesis correlates well with the increased capacity of liver nuclei to incorporate radioactive nucleoside triphosphates into RNA in vitro[1,4-6]. We have reported earlier that template efficiency of liver chromatin (though not activity of RNA polymerase as isolated free of template DNA) was significantly stimulated as a result of total-body radiation exposure[1]. Since the ability of liver nuclei to incorporate radioactive precursors into RNA was increased to a much higher extent (187% over non-irradiated control) than the increase in template efficiency of chromatin (about 51% over non-irradiated control)[1], it appeared that some other factors may be involved in the activation of nuclei. We have now obtained evidence which indicates that the liver nuclei could have become more permeable to nucleoside triphosphate precursors as a result of whole-body radiation exposure. Such change in permeability was not discerned if nuclei were irradiated in vitro.

Materials and methods. Wistar strain rats, each weighing between 150 and 160 g fed on a laboratory diet, were used. Rats were given a single whole-body dose of 1000 R of X-irradiation by housing them in groups of 3, in a 3-place perspex container located at the distance of 50 cm from 250 kV X-ray generator (Seimen's Stabilipan). The latter was operated at 15 mA tube current with added filter 2 mm Al. The dose rate was 100 R/min.

The procedures for isolation and purification of liver nuclei and assay for Mg^{++}-dependent and $Mn^{++}/(NH_4)_2SO_4$-dependent RNA polymerase reactions were carried out according to Widnell and Tata[7]. For in vitro irradiation of liver nuclei, isolated nuclei were suspended in 0.9% saline (≡3 mg DNA/ml) and irradiated by the X-ray generator at 0–2 °C.

Uptake of ^{3}H-UTP by isolated nuclei in vitro was studied by incubating the nuclear suspension (≡0.3 mg DNA) separately in the reaction mixtures (in 0.5 ml final volume) for assays of Mg^{++}-dependent and $Mn^{++}/(NH_4)_2SO_4$-dependent RNA polymerase activities with the following changes: Instead of the 4 nucleoside triphosphates, 0.3 µmoles of ^{3}H-UTP (7.5 µCi/mmole) and 15 µg actinomycin D were included in each reaction mixture. Actinomycin D was included to suppress synthesis of RNA, if any, from endogenous ribonucleoside triphosphates. The incubations were stopped by placing the assay tubes immediate-

Table 1. RNA polymerisation capacities of liver nuclei: effect of whole-body X-irradiation

	Amount of RNA synthesized	
	Mg^{++}-dependent*	$Mn^{++}/(NH_4)_2SO_4$-dependent**
Non-irradiated	256 ± 13	1078 ± 19
Irradiated (1000 R) h		
4	306 ± 11	1150 ± 23
12	410 ± 14	1243 ± 9
18	724 ± 25	1490 ± 11
36	287 ± 16	1120 ± 15

*pmoles of AMP incorporated into RNA/15 min/mg DNA for Mg^{++}-dependent reaction. **pmole of AMP incorporated into RNA/45 min/mg DNA for $Mn^{++}/(NH_4)_2SO_4$-dependent reaction. Both Mg^{++}-dependent and $Mn^{++}/(NH_4)_2SO_4$-dependent assay systems contained in a volume of 0.5 ml: 0.02 µmoles of ^{14}C-ATP (sp.act. 27.2 mCi/mmole) and nuclei equivalent to 0.3 mg DNA. Other components are as described by Widnell and Tata[7]. Each value is average of 3 determinations on nuclei from pooled livers (2 rats per group) ± SEM.

Table 2. Uptake of ^3H-uridine triphosphate by isolated rat liver nuclei in vitro: effect of whole-body X-irradiation

	^3H-UTP taken up by nuclei from reaction mixtures containing	
	Mg^{++} (pmoles)	$Mn^{++}/(NH_4)_2SO_4$ (pmoles)
Non-irradiated	34.60 ± 5.9	64.60 ± 15.0
Irradiated (1000 R) h		
4	40.30 ± 6.8	64.90 ± 10.6
12	44.60 ± 6.0	66.60 ± 12.6
18	68.80 ± 7.2	68.20 ± 18.0
36	37.30 ± 5.7	65.40 ± 5.0

Nuclei (equivalent to 0.3 mg DNA) were incubated at 37 °C for 10 min in reaction mixtures (final volume 0.5 ml) used for assaying Mg^{++}-dependent and $Mn^{++}/(NH_4)_2SO_4$-dependent RNA polymerisation, except that ^3H-UTP (0.3 µmoles) and actinomycin-D (15 µg) were included in place of ^{14}C-ATP, GTP, UTP and CTP. Other details are as described in the methods. Each value is average of 3 determinations on nuclei from pooled livers (2 rats per group) ±SEM.

Table 3. RNA polymerisation capacities of liver nuclei at various times following whole-body X-irradiation (corrected for changes in permeability of liver nuclei to nucleoside triphosphate)

	Amount of RNA synthesized	
	Mg^{++}-dependent*	$Mn^{++}/(NH_4)_2SO_4$-dependent**
Non-irradiated	256.0	1078
Irradiated (1000 R) h		
4	263.4	1143
12	318.7	1206
18	364.5	1410
36	266.9	1106

*pmoles of AMP incorporated into RNA/15 min/mg DNA for Mg^{++}-dependent reaction. **pmoles of AMP incorporated into RNA/45 min/mg DNA for $Mn^{++}/(NH_4)_2SO_4$-dependent reaction. Corrected RNA polymerisation capacities were calculated as follows:

$$\text{Amount of RNA synthesized in liver nuclei from irradiated rats (table 1)} \times \frac{\text{UTP uptake by liver nuclei from non-irradiated rats (table 2)}}{\text{UTP uptake by liver nuclei from irradiated rats (table 2)}}$$

ly in an ice-bath. The nuclei were collected by centrifugation at 1600 × g, resuspended in cold 0.25 M sucrose containing 0.1 M unlabelled UTP, recentrifuged and counted for radioactivity.

Results and discussion. Widnell and Tata[7] have demonstrated that in isolated rat liver nuclei, RNA polymerase activity can be resolved into 2 reactions, one manifests itself in the presence of Mg^{++} ions and low ionic strength yielding a product similar to ribosomal RNA, and the other which is predominant in the presence of Mn^{++} ions and high concentration of salt, such as 0.4 M $(NH_4)_2SO_4$, the product of which is DNA-like RNA. As seen in table 1, total-body exposure to X-rays results in a marked elevation in both the Mg^{++}-dependent and $Mn^{++}/(NH_4)_2SO_4$-dependent RNA polymerisation activites in vitro in isolated liver nuclei. The maximum increases were found at 18 h post-irradiation, being 187% over non-irradiated control for Mg^{++}-dependent reaction and 36% over non-irradiated control for $Mn^{++}/(NH_4)_2SO_4$-dependent reaction.

The observed increase in the capacities of nuclei to synthesise (i.e. label) RNA seems to be related to activation of transcription machinery, but, in addition, also to factors such as increased permeability of liver nuclei to take up nucleoside triphosphate precursors. The possible whole-body radiation effect on the permeability of liver nuclei was hence investigated by studying the uptake of ^3H-UTP in the presence of actinomycin D added to prevent RNA polymerisation. The results are shown in table 2. It will be seen that the uptake of ^3H-UTP by liver nuclei from non-irradiated rats is much higher in the presence of $Mn^{++}/(NH_4)_2SO_4$ than in the presence of Mg^{++}. This may be due to the higher concentration of $(NH_4)_2SO_4$ which may render nuclei more permeable to nucleoside triphosphate precursors. Liver nuclei from irradiated rats were far more efficient in ^3H-UTP uptake in the presence of Mg^{++} than those from non-irradiated rats. There was, however, no significant difference between liver nuclei derived from the 2 groups of rats in respect of ^3H-UTP uptake studied in the presence of $Mn^{++}/(NH_4)_2SO_4$. Possibly the increased permeability caused by high concentration of $(NH_4)_2SO_4$ could have obliterated any differences in permeability in liver nuclei derived from the 2 groups of rats. Notwithstanding their increased permeability, nuclei of irradiated rats are more efficient in RNA polymerisation as compared to those from non-irradiated controls. This is clearly seen from table 3 in which the extent of RNA polymerisation by liver nuclei from irradiated rats is calculated, taking into account the changes in permeability of the nuclei to ribonucleoside triphosphates.

Table 4. Uptake of ^3H-uridine triphosphate by isolated rat liver nuclei in vitro: effect of in vitro X-irradiation

	^3H-UTP taken up by nuclei from reaction mixtures containing	
	Mg^{++} (pmoles)	$Mn^{++}/(NH_4)_2SO_4$ (pmoles)
Non-irradiated	30.7 ± 5.5	58.7 ± 10.5
Irradiated		
3000 R	30.6 ± 6.9	60.1 ± 12.4
5000 R	31.3 ± 5.2	59.4 ± 9.4

Nuclei (equivalent to 0.3 mg DNA) were incubated at 37 °C for 10 min in reaction mixtures (final volume 0.5 ml) used for assaying Mg^{++}-dependent and $Mn^{++}/(NH_4)_2SO_4$-dependent RNA polymerisation, except that ^3H-UTP (0.3 µmoles) and actinomycin-D (15 µg) were included in place of ^{14}C-ATP, GTP, UTP and CTP. Other details are as described in the methods. Each value is average of 3 determinations on nuclei from pooled livers (2 rats per group) ± SEM.

The activation of transcription machinery of the liver following whole-body X-irradiation does not seem to be due to the effect of irradiation directly on the liver itself, but possibly arises as a result of adrenal mechanisms[4-6]. Our results have further indicated that irradiation of liver nuclei in vitro does not enhance or decrease RNA polymerisation capacities (M.N. Subba Rao, M.S. Netrawali and D.S. Pradhan unpublished observation). As seen in table 4 in vitro irradiation of liver nuclei likewise does not change the permeability of nuclei to nucleoside triphosphates, even at the doses as high as 5000 R. This would mean that the increased permeability of nuclei to ^{3}H-UTP could also have arisen from indirect mechanisms.

It has been shown that the increase in the ability of liver nuclei from irradiated rats to synthesis RNA in vivo could be mediated through glucocorticosteroids known to be released in greater amounts after whole-body radiation exposure probably through hypothalamus-pituitary-adrenal axes[8-11]. The whole-body radiation-induced changes in permeability of liver nuclei could hence be conceived as the consequences of increased release of glucocorticosteroids. Our results showing that irradiation in vitro fails to elicit changes in permeability of nuclei seem to support this contention. Indeed, a study reported by Sekeris and coworkers seems to suggest that cortisol added in vitro may change permeability of liver nuclei to iodoacetic acid[12].

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Slavery in the subfamily Dolichoderinae (F. Formicidae) and its ecological consequences

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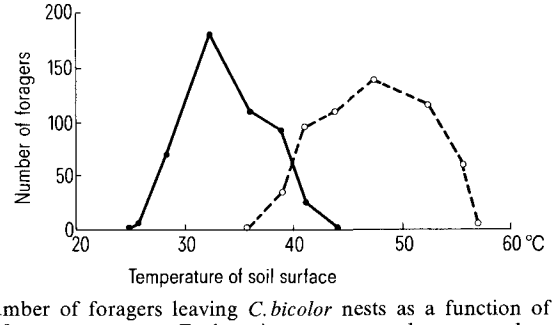
Summary. Evidence of slave-making habits in 2 species of the subfamily Dolichoderinae has been found in arid habitats of western North America. The enslaved species are of the subfamilies Myrmicinae and Formicinae. In previously reported cases of slavery in ants, both the slave-making and enslaved species are of the subfamily Formicinae. In the 2 new cases of slavery reported here, presence of slaves of another species in a colony significantly increases the breadth of diet and/or the range of temperatures at which the colony forages.

Certain species of ants are known to capture pupae from the nests of other ant species, return them to their own nests, and allow them to emerge as workers. In known cases of slavery, both the slave-makers and the enslaved species are within the subfamily Formicinae. In my studies of the ecology of ants, I have observed 2 possible cases of slavery: in both, the slave-making species is in the subfamily Dolichoderinae. The enslaved species are of different subfamilies, Myrmicinae in one case and Formicinae in the other. The present study describes the evidence for slavery in this new subfamily and the ecological changes resulting from the inferred slavery systems.

The 1st case of slavery reported here is that of *Conomyrma bicolor* using workers of *Myrmecocystus kennedyi* as slaves. The colonies were observed in the Mojave Desert of southwestern United States, at an elevation of 825 m. While taking ecological measurements on what were believed to be compound nests of the 2 species, I observed slave raids by foragers of *C. bicolor* on colonies of *M. kennedyi*. These raids developed in several of the 'compound' nests at about 16.30 h on the afternoons of 19–23 July. The soil surface temperature at this time was 37 °C; the ambient temperature 32.6 °C. Initially, *C. bicolor* workers began to emerge from their nests in unusually large numbers and to mill around the nest entrance for about 30 min. About 300 workers per colony were present and all foraging ceased. Workers of each colony then formed into a loose column and traveled up to 10 m from their nest to enter the nest of *M. kennedyi*. The *M. kennedyi* workers gave little resistance and soon thereafter, *C. bicolor* workers began to return to their own nest, each carrying a pupa. This continued until sunset each day, at which time the soil surface temperature was around 29 °C and the ambient temperature 27 °C. During the latter part of the raid, *M. kennedyi* workers were seen leaving their raided nests, some carrying pupae.

The 2nd case of slavery was observed at a study site located in sagebrush habitat (elevation 1524 m) in the Great Basin Desert of western United States (Arizona). On the mornings of 27–30 June, workers of *Conomyrma insana* colonies were seen to travel in loose columns to the nests of



Number of foragers leaving *C. bicolor* nests as a function of soil surface temperature. Each point represents the mean observed, during a 10-min interval, for 4 colonies. Solid line represents *C. bicolor* foragers; dashed line represents *M. kennedyi* foragers.