

Antibacterial activity in vitro of 2-amino-4(2-ethynyl-1-methyl-5-nitroimidazole)-pyrimidine (F4)

Microorganism	Tryptic soy broth	Tryptose phosphate broth	Tryptic soy broth + 5% fetal calf serum	Tryptic soy broth + 10% fetal calf serum
<i>Staphylococcus aureus</i> ^a	1	25	—	—
<i>Staphylococcus aureus</i> 168	25	—	—	—
<i>Staphylococcus aureus</i> B-ICI	5	—	—	—
<i>Streptococcus pyogenes</i> ^a	0.5	—	—	—
<i>Streptococcus pyogenes</i> ISM 68/237	2.5	—	—	—
<i>Streptococcus viridans</i> ^a	—	10	—	—
<i>Streptococcus faecalis</i> ^a	50	—	—	—
<i>Diplococcus pneumoniae</i> ^a	5	—	—	—
<i>Bacillus cereus</i> ATCC 11778	2.5	2.5	—	—
<i>Escherichia coli</i> K12	10	—	—	—
<i>Salmonella enteritidis</i> ISM 66/33	5	10	10	10
<i>Salmonella paratyphi</i> A ISM 66/18	2.5	5	—	—
<i>Salmonella paratyphi</i> B ISM 66/19	2.5	1	2.5	2.5
<i>Salmonella typhi</i> 0-901	5	—	—	—
<i>Klebsiella pneumoniae</i> ATCC 10031	2.5	1	2.5	2.5
<i>Proteus vulgaris</i> ATCC 6380	25	50	—	—
<i>Proteus mirabilis</i> ^b	10	50	50	50
<i>Pseudomonas aeruginosa</i> ^b	50	50	—	—
<i>Shigella dysenteriae</i> Madsen	25	—	—	—

Minimal inhibiting concentrations (μg/ml), 12 h. The data are the mean of at least 4 experiments. ^a From the Institute of Hygiene of the University of Modena. ^b From the Institute of Microbiology of the University of Parma.

good activity just against those bacteria which are more commonly associated with *Trichomonas vaginalis*.

Riassunto. Il 2-amino-4(2-etinil-1-metil)5-nitroimidazolo-pirimidina (F4), dotato di attività antitricomoniasica almeno pari a quella del metronidazolo, a differenza di questo si dimostra capace di svolgere in vitro una rimar-

chevole attività inibente verso lo sviluppo di germi sia Gram-positivi sia Gram-negativi.

A. BERTOLINI, M. CASTELLI and R. POGGIOLI

Cattedra di Chemioterapia, Istituto di Farmacologia, Via G. Campi 287, I-41100 Modena (Italy), 2 July 1973.

The Probable Significance of the Differential Occurrence of Protein in Various Castes of the Termite *Odontotermes assmuthi* (Isoptera: Termitidae)

The metabolic significance of fat as fuel and protein for tissue building is well known. Recently, BASALINGAPPA and HEGDE (unpublished) found that chicks fed with dried queens of the termite *Odontotermes assmuthi* showed better growth compared with controls. While there have been many studies¹⁻⁴ including high amounts

of fat in termites, our knowledge on protein content in different termite castes is too meagre. The only available report is from TIHON¹ who has stated that protein amounts to 36% in unspecified termite alates. The present note reports the percentage of protein in different castes of the termite *O. assmuthi*.

Materials and methods. Various castes of the termite *O. assmuthi* were collected in the vicinity of Karnatak University Campus, Dharwar (Mysore State, India). They were dried at 100°C for 24 h and fat was extracted following the method of FOLCH et al.⁵. The fat-free dry residue was used for the estimation of protein⁶.

Results and discussion. The Table shows the percent of protein on dry basis in different castes of the termite *O. assmuthi*. The results indicate that the amount of protein is highest in queens (65.7%), which is more or less double the protein value (37%) of female alates from

Mean percent protein on dry basis in different castes of the termite *Odontotermes assmuthi*

No.	Castes	% Protein by dry weight (average of 5 readings)
1.	Potential reproductive forms	
	a) Male alates	47.9 ± 0.83
	b) Female alates	37.4 ± 1.81
2.	Functional reproductive forms	
	a) Kings	47.6 ± 7.78
	b) Queens	65.7 ± 4.75
3.	Active (neutral) forms	
	a) Workers	33.5 ± 3.02
	b) Soldiers	44.5 ± 4.57
4.	Undifferentiated instars	29.76 ± 1.80

¹ L. TIHON, Bull. Agric. Congo Belg. 37, 865 (1946).

² S. H. W. CMELIK, J. Insect Physiol., 15, 839 (1969).

³ S. H. W. CMELIK, J. Insect Physiol. 15, 1481 (1969).

⁴ S. BASALINGAPPA, J. Anim. Morph. Physiol. 2, 106 (1970).

⁵ J. FOLCH, M. LEES and G. H. S. STAINLEY, J. biol. Chem. 226, 497 (1957).

⁶ O. H. LOWRY, N. J. ROSEBROUGH, A. L. FARR and R. J. RANDALL, J. biol. Chem. 193, 265 (1951).

which the queens develop. The reasons suspected for such a high protein content in queens are as following: 1. the queens are sufficiently fed with proteinaceous food by workers; 2. they are much bloated and physogastric, weighing from 20 to 200 times more than the female alates; 3. the ovaries being filled with an enormous number of proteinaceous eggs comprise the bulk of the body weight of the physogastric queen.

The kings, which are developed from male alates, show the same 47% protein as male alates. The reason may be presumed to be that the kings do not undergo any marked change, as do the queens. Among neuter castes, the soldiers have almost an equal amount of protein as potential sexual castes (Table). This is suspected to be for their proteinaceous salivary fluid which fills the salivary receptacles which in turn occupy the major portion of the abdomen in soldiers. In support of this opinion regarding the content of salivary fluid, BOUILLON⁷, while studying the termites of Ethiopian region, mentions that the saliva doubtless contains proteinaceous material. In conclusion, the differential occurrence of protein in various castes and undifferentiated instars of the termite *O. assmuthi* is solely dependent upon the different roles played by each caste and as due to the regulated feeding.

Résumé. L'évaluation de la somme de protéine contenue dans les représentants des diverses castes du termite *Odontotermes assmuthi* donne les résultats suivants: 1. Les formes reproductrices potentielles, mâles et femelles, ont des quantités significatives (47,9 et 37%) que requièrent leur activité reproductrice. 2. Les rois issus de mâles ailés n'atteignant leur pourcentage maximum de protéine qu'à l'âge adulte. 3. Les reines en augmentent leur quantité (à 65,7%) en proportion de leur intense production d'œufs. 4. Les ouvriers et les soldats soumis à des tâches ardues contiennent beaucoup de protéine (33,5 et 44,5%).

S. BASALINGAPPA⁸ and S. N. HEGDE

Department of Zoology, Karnatak University,
Dharwar-580003 (Mysore State, India), 19 June 1973.

⁷ A. BOUILLON, in *Biology of Termites*, (Eds. K. KRISHNA and F. M. WEESNER; Academic Press, New York 1970), vol. 2, p. 153.

⁸ Acknowledgment. The authors are indebted to Professor C.J. GEORGE (C.S.I.R.) for his valuable guidance and thanks to Professor M. APPASWAMY RAO, for the facilities provided.

The Membrane Expansion Theory of Anesthesia: Direct Evidence Using Ethanol and a High-Precision Density Meter

The membrane expansion theory of anesthesia¹ states that anesthetics and other nerve-blocking drugs adsorb to hydrophobic regions of excitable membranes²⁻⁵, expanding the hydrophobic regions of membrane proteins⁶⁻¹² and thus blocking the ionic conductance channels underlying nerve cell action potentials.

Surgical concentrations of general anesthetics do expand the membrane area of erythrocytes by about 0.4%, while the volume of the anesthetic which occupies the membrane under these conditions is only 0.02% or less^{1,7}. Since the membrane expansion is roughly 20 times the occupying volume of drug in the membrane phase, it has been suggested that extensive conformation changes in membrane proteins may be involved¹. Although it is known that anesthetics expand the area of erythrocyte membranes, this report provides the first direct evidence that anesthetics also expand the specific volume of such membranes.

Erythrocyte membranes and guinea-pig brain synaptosomes¹³ were suspended in 10 mM sodium phosphate buffer ([H⁺] = 40 nM). The final dry weight of the membranes ranged from 0.2 to 1 g per 100 ml of suspension. Using a precision density meter¹⁴ at 25 ± 0.01°C, the density of the dry membrane (d_m) was obtained by eq. 1 (see appendix):

$$d_m = \frac{W \times d_{ms}}{f(d_b - d_{ms}) + h(1 - d_{ms}) + W} \dots \dots \dots (1),$$

where W is the dry weight of the membranes (in g dry membrane per ml of suspension), where d_{ms} and d_b are the measured densities of the membrane suspension and the buffer solution, respectively, in the presence of varying concentrations of drug, where h is the fraction of membrane-associated water (i.e. non-solvent water) in the suspension (having dimensions of ml water per ml of suspension), and where f is the fraction of buffer medium in the suspension (in units of ml buffer medium per ml suspension). The amount of membrane-associated water

is generally 0.3 ml per g of dry membrane¹⁵; that is, $h = 0.3 W$. The value for f was then taken as $1 - W - h$.

Ethanol lowered the density of the membrane suspension disproportionately more than that of the buffer solution, qualitatively indicating that the density of the biological membranes decreased in the presence of the drug. The values for d_b and d_{ms} , respectively, (in units of g/cm³, with an error of $\pm 1.5 \times 10^{-6}$ g/cm³) in a typical experiment were: 0.997950 and 0.998649 for 0 M ethanol

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⁶ S. ROTH and P. SEEMAN, *Nature, Lond.* 231, 284 (1971).

⁷ P. SEEMAN and S. ROTH, *Biochim. biophys. Acta* 255, 171 (1972).

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⁹ M. J. LEVER, K. W. MILLER, W. D. M. PATON and E. B. SMITH, *Nature, Lond.* 231, 368 (1971).

¹⁰ K. W. MILLER, W. D. M. PATON, R. A. SMITH and E. B. SMITH, *Molec. Pharmac.* 9, 131 (1973).

¹¹ C. S. SPYROPOULOS, *J. gen. Physiol.* 40, 849 (1957).

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¹⁴ O. KRATKY, H. LEOPOLD and H. STABINGER, *Z. angew. Physiol.* 27, 273 (1969), technical translation 1583 of Nat. Res. Council (Ottawa, Canada). The instrument used was a precision density meter (Model DMA 02C) made by Anton Paar, K.G., Graz, Austria.

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