ISSN 0300-9254

Appropriate Technology Service
121, POINT-PEDRO ROAD
NALLUR, JAFFNA
No. 1853

JOURNAL OF THE NATIONAL SCIENCE COUNCIL OF SRI LANKA

VOLUME 9 NO. 1

June 1981

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Journal of the National Science Council of Sri Lanka

EDITORIAL BOARD: R. P Jayewardene (Chairman)
B. A. Abeywickrema
G. A. Dissanaike
P. D. Gunatilaka
T. W. Herath
W. P. Jayasekera
H. N. S. Karunatilake
N. D. W. Lionel
R. S. Ramakrishna
Y. D. A. Senanayake
S. Wijesundera
Nimala Amarasuriya (Editor)

PUBLICATION: One volume of two issues (June and December) is published annually by the National Science Council of Sri Lanka.

Subscription Annual subscription–Foreign $ 27.00; Local Rs. 45.00.
Accepted on a calendar year basis. Rates include postage.

Single issues–Foreign $ 13. 50; Local Rs. 27.50 each
Back issues –Foreign $ 5.00; Local Rs. 12.50 each
Rates include postage.

Payment must accompany all orders. Remittances in favour of National Science Council of Sri Lanka.

Change of address notice is required 4 weeks before issue date. Orders and all correspondence relating to them should be sent to the **Accountant** National Science Council of Sri Lanka at the address given below.

Manuscripts Research Papers, Reviews and Short Communications in all fields of Science and Technology in Sinhala, Tamil and English may be submitted for editorial consideration. Manuscripts should conform to the style adopted in this issue. For instructions as to preparation of papers see inset at back of this issue. Separates of General Instructions and special Instructions in Chemical, Physical and Medical Sciences may be had on application to the Secretary, Editorial Board at the address given below.

No responsibility is assumed by the National Science Council of Sri Lanka for statements and opinions expressed by the contributors to this Journal.

Manuscripts and all correspondence relating to them should be sent to the **Secretary**, Editorial Board, Journal of the National Science Council of Sri Lanka, 47/5, Maitland Place, Colombo 7, SRI LANKA.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Appropriate Technology Service
121, POINT ROAD
NALLUR, JAFFNA
No. 853

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1)

Preliminary Studies on the Alginic Acid and Agar contents of some Marine Algae

INDRANEE ARUMUGAM, A. SIVAPALAN AND K. THEIVENDIRARAJAH

Department of Botany, University of Jaffna, Thirunelvely, Sri Lanka.

(*Date of receipt* : 29 May 1979)

(*Date of acceptance* : 03 November 1980)

Abstract: Different methods of extraction of alginic acid from brown algae were tried out and it was found that treating with Na_2Co_3 followed by bleaching powder gave alginic acid of better quality. Of the several species of brown algae studied *Cystoseira triquetra* (L) J. Agardh and *Turbinaria conoides* (Kuetzing) were found to contain the greatest amounts of alginic acid. Investigations further showed that there was a marked seasonal variation in the content of alginic acid in these algae, highest being during the months of May to July. Extraction of agar was carried out in six different species of red algae and it was found that species of *Gelidium* (Lamouroux) *Hypnea* (Lamouroux) and *Gracilaria* (Greville) could be used for extracting agar. The amount of agar in these algae varied from 16·2% to 50%. There is a marked seasonal variation in the agar content in these algae. The content of agar was found to be high in these algae during January.

1. Introduction

Alginic acid and agar are two important commercial polysaccharides that could be obtained from marine algae. Algin is usually obtained from some brown algae and the alginic acid producing algae are generally called alginophytes. Similarly agar can be obtained from some red algae and the agar producing algae are called agarophytes.

Both alginic acid and agar are extensively used in Sri Lanka and annually large quantities of these products are imported. Agar is mainly used in confectionaries as a substitute for gelatine. It is also used as a culture medium for the growth of micro-organisms like bacteria and fungi. Alginic acid as its sodium salt is extensively used in textile industry.

No systematic studies have been made on the content of alginic acid and agar present in the marine species found in Sri Lanka. The present investigations were undertaken to estimate quantitatively the amounts of these substances in a number of marine algae found along the coast of Mandaitivu in the Jaffna Peninsula, described by Durairatnam[1]. Mandaitivu, is an island situated on the South West side of the Jaffna peninsula. It is about five miles from the main land and there is a rich flora of alginophytes and agarophytes along the coast of Mandaitivu.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

2. Methods of Study and Results

2. 1. Extraction of alginic acid ;

Different methods used by Rao and Modi[2] and Rao *et al*[3] were tried for the extraction of alginic acid from brown algae.

2. 1. 1. – Extracted with sodium carbonate and precipitated with dilute hydrochloric acid.

2. 1. 2. – Extracted with sodium hydroxide and precipitated with dilute hydrochloric acid.

In these two methods the extract contains sodium alginate. When dilute hydrochloric acid is added precipitation of alginic acid takes place. Here the product is soft in texture and dark brown in colour. Since the colour and texture of the product obtained by these two methods were not satisfactory another method was tried.

2. 1. 3. – Extracted with $Na_2 CO_3$, precipitated with bleaching powder, added dilute HCl, until effervescence ceases. Here calcium alginate and calcium carbonate precipitate and then the latter is removed completely. In this method the alginic acid obtained is rubbery in texture and white in colour. Therefore the ultimate method adopted for the routine estimation of alginic acid content is as follows:

20 ml of 6% sodium carbonate solution of pH around 9 is added to 0. 5g dry wt of the algal material and kept at 50°C for nearly 3 hours for the material to become a pulp. Extract is filtered through a muslin cloth, saturated solution of bleaching powder is added until complete precipitation occurs. Dilute hydrochloric acid is added until CO_2 effervescence ceases, product filtered, washed in water,dried at 50°C and weighed.

This method is very economical because of the cheap reagents used.

A quantitative study on the amount of alginic acid present in the following marine algae such as *Padina* (Adanson), *Cystoseira triquetra* (L). J. Ag., *Turbinaria conoides* (Kuetzing), *Cystophyllum* (J. Agardh), *Turbinaria ornata* (J. Agardh), *Hydroclathrus* (Bory), *Sargassum polycystum* (C. Ag), *Sargassum whightii* (Greville), *Sargassum cervicone*, (Greville), *Stoechospermum* (Kuetzing), *Dictyota* (Lamouroux) and *Sargassum tenerrimum* (J. Ag.) was carried out periodically using duplicate samples at about $1\frac{1}{2}$ to 2 months interval beginning from October 1977. The results of this analysis is given in Table I.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 1 – Alginic acid content (percent dryweight) of different brown algae collected from Mandaitivu during the period of survey.

Species	October 1977	December 1977	January 1978	March 1978	May 1978	July 1978	August 1978
Padina ..	10%	8%	4%	8%	14%	4%	—
	8%	10%	4%	10%	14%	4%	—
Dictyota ..	10%	—	14%	—	—	—	—
	12%	—	16%	—	—	—	—
Stoechospermum ..	—	—	—	—	—	14%	8%
						16%	8%
Cystoseira ..	—	20%	14%	24%	33%	32%	18%
	—	18%	16%	26%	32%	28%	16%
Cystophyllum ..	14%	10%	10%	18%	—	10%	10%
	16%	10%	12%	18%	—	10%	10%
Turbinaria conoides ..	28%	20%	12%	—	—	—	8%
	24%	20%	12%	—	—	—	8%
Turbinaria ornata ..	—	—	6%	18%	20%	28%	8%
	—	—	6%	18%	20%	28%	10%
Hydroclathrus ..	—	—	12%	—	—	—	—
	—	—	14%	—	—	—	—
Sargassum polycystem ..	20%	14%	12%	—	22%	24%	—
	20%	14%	12%	—	22%	28%	—
Sargassum whightii ..	16%	12%	—	—	—	—	12%
	14%	12%	—	—	—	—	10%
Sargassum cervicone ..	—	—	—	—	—	12%	—
	—	—	—	—	—	14%	—
Sargassum tenerrimum ..	—	—	14%	22%	28%	—	—
	—	—	16%	22%	30%	—	—

(—) indicates that the species is absent.

Results show that there is significant difference in the alginic acid content of different species tested. Also a marked seasonal variation is present in all species. Percentage alginic acid is high during the period May to July, and is low during December to January. Of the species tested *Cystoseira triquetra* contained the greatest amount of alginic acid. However there was no difference in the texture of the alginic acid extracted from the different species.

2. 2. Moisture content of different alginophytes.

The percentage moisture in the thallus of the different alginophytes that have been used in this study was determined in order to find out whether there is any relationship between the alginic acid content and moisture content. Moisture

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

content was determined by drying 10g of freshly collected algae, after surface blotting, in an oven at 80°C until constant weight is attained. Percentage coisture ,of different species during the period of survey is given in Table 2.

Table 2 – Percentage moisture (percent fresh weight) of the thalli of alginophytes used in this study

Species.	October 1977	December 1977	January 1978	March 1978	May 1978	July 1978	August 1978
Padina	58%	66·6%	61·5%	64·3%	61·3%	68·7%	—
Dictyota	66%	—	64%	—	—	—	—
Stoechospermum	—	—	—	—	—	76·2%	74·5%
Cystoseira	—	84·4%	78·3%	80·8%	79·2%	81·5%	83·5%
Cystophyllum	75%	78·5%	73·6%	68·7%	—	76·2%	73·1%
Turbinaria conoides	75·22%	75·22%	72·22%	—	—	—	71·3%
Turbinaria ornata	—	—	66%	63·2%	61·4%	75·1%	76·2%
Hydroclathrus	—	—	90·3%	—	—	—	—
Sargassum polycystum	79·4%	72·2%	71·0%	—	70%	72·2%	—
Sargassum whightii	70·6%	72·5%	—	—	—	—	76%
Sargassum cervicone	—	—	—	—	—	72·2%	—
Sargassum tenerrimum	—	—	68·4%	64·3%	62·8%	—	—

(—) indicates that the species is absent·

The results indicate that there is no significant correlation between alginie acid content and moisture content. There is variation among species in the moisture content, but there is no marked seasonal variation in any of the alginophytes tested.

2. 3. Extraction of Agar.

Agar was extracted according to the method described by J. F. Wood.[4] 2 g of dry sea weed is soaked in 50ml water overnight. Depending upon the species this is either boiled or autoclaved after adding 3–5 drops of dilute hydrochloric acid. The pH of the extraction medium is around 6. When the tissue turns into a pulp it is filtered, allowed to set, sundried and weighed. Extraction of agar was tried on five different species of red algae namely *Gelidium* (Lamouroux) *Gracilaria lichenoides* (L) Harvey, *Hypnea* (Lamouroux), *Acanthophora* (Lamouroux) and *Laurencia* (Lamouroux). Routine estimation was carried out periodically using duplicate samples to see whether there is any fluctuation of agar content. Results of this analysis is given in Table 3.

Results show that of the species tested *Gelidium*, *Gracilaria lichenoides* and *Hypnea* are quite suitable for the extraction of agar while *Acanthophora* and *Laurencia* cannot be used for this purpose. Seasonal variation is present in the agar content of all species tested. Generally agar content seems to be high during January and low during May.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 3 – Agar content (percent dry weight) of different red algae collected from Mandaitivu during the period of study.

Species.		October 1977	December 1977	January 1978	March 1978	May 1978	July 1978	August 1978
Hypnea	1	36·18%	—	46·1%	—	16·3%	47·5%	—
	2	35·8%	—	46·28%	—	16·1%	47·45%	—
	Av	36%	—	46·2%	—	16·2%	47·5%	—
Gracilaria lichenoides	1	36·3%	—	49·9%	35%	28·5%	32·2%	26·6%
	2	36·7%	—	50·1%	35·1%	26·4%	32·75%	26·4%
	Av	36·5%	—	50%	35%	27·5%	32·5%	26·5%
Gelidium	1	40%	23·2%	39·51%	32·8%	22%	24·5%	21%
	2	40%	22·75%	39·48%	31·16%	23·1%	25·4%	22%
	Av	40%	23%	39·5%	32%	22·5%	25%	21·5%
Acanthophora	1	—	—	—	—	—	—	—
	2	—	—	—	—	—	—	—
	Av	—	—	—	—	—	—	—
Laurencia	1	—	—	—	—	—	—	—
	2	—	—	—	—	—	—	—
	Av	—	—	—	—	—	—	—

Hypnea and G. *lichenoides* (—) indicates absence of species.
Acanthophora and *Laurencia* (—) indicates that there is no extractable agar in them.

2. 4. Moisture content of agarophytes.

Percentage moisture of the different agarophytes used in this study is given in Table 4. The moisture content of the agarophytes was determined as described in 2·2. There seems to be no correlation between agar content and moisture content of these algae.

Table 4 – Moisture content per cent fresh weight of different agarophytes used in this study

Gracilaria lichenoides	..	..	—	86·5%
Gelidium	..	..	—	70%
Hypnea	..	..	—	87·1%
Acanthophora	..	..	—	88·4%
Laurencia	.	..	—	88·1%

2. 5. Abundance of the alginophytes and agarophytes.

An eye estimation of the abundance of different alginophytes and agarophytes was carried out during different months from October 1977 to August 1978. Results are given in Tables 5 and 6.

Results indicate that certain alginophytes and agarophytes such as *S. tenerrimum*, *Acanthophora*, *Laurencia*, *Gelidium* are present throughout the year although the relative abundance during different months vary. Others such as *Cystoseira*, *Cystophyllum*, *S. polycystum* are present during most of the months and are completely absent during some periods of the year.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 5 – Relative abundance of alginophytes found at Mandaitivu during different periods of the year.

Species.	October 1977	December 1977	January 1978	March 1978	May 1978	July 1978	August 1978
Padina ..	+	++	++++	++	+++	+	—
Dictyota ..	++	—	+	—	—	—	—
Stoechospermum ..	—	—	—	—	—	+++	++++
Cystoseira ..	—	++++	+++	+++	+++	++++	+
Cystophyllum ..	+++	++++	++++	+++++++	—	++++	+
Turbinaria conoides ..	++++	+++++	++	—	—	—	++++
Turbinaria ornata ..	—	—	+++	—	—	+++	++
Hydroclathrus ..	—	—	++	—	—	—	—
Sargassum polycystum ..	++	+++	++++++	—	+++	++++	—
Sargassum whightii ..	+++++	++	—	—	—	—	++++
Sargassum cervicone ..	—	—	—	—	—	++++++	—
Sargassum tenerimum ..	—	—	++	++++	+++	—	—

(—) Indicates absence of the species.

+++++++ indicates the most abundant species.

\+ indicates that the species is present in trace amounts.

The population of the other species lie inbetween these two.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 6 – Relative abundance of red algae found at Mandaitivu.

Species	Oct. 1977	Dec. 1977	Jan. 1978	Mar. 1978	May 1978	July 1978	Aug. 1978
Hypnea	+ +	—	+ + + +	—	+ +	+ + + + + +	—
Gracilaria lichenoides	+ + + + +	—	+ +	+ +	+ + +	+ + +	+ + + + +
Gelidium	+ +	+ +	+ +	+ +	+ + +	+	+
Acanthophora	+ + + +	+	+	+ + + +	+ + + +	+ + +	+ + + + + + +
Laurencia	+ + + +	+ + +	+ + +	+ + + +	+ + + +	+ + +	+ + + + + + +

+ + + + + + + — The most abundant species.
\+ Species present in trace amounts
The relative abundance of the other species lie inbetween these two.
(—) indicates absence of the species.

Species such as *Hydroclathrus, S. whightii, S. cervicone, T. ornata* appear only during certain months of the year.

3. Discussion.

Alginic acid and agar could be produced in small cottage industries in Sri Lanka since the natural resources necessary for this purpose are available in plenty. The crude agar obtained by the method of extraction mentioned here could be utilised in certain microbiological work. The crude agar preparation sets well at 1. 5 percent. This setting property is not affected at pH ranging from 3 to 9. The alginic acid obtained in our studies is a white pulp of gelatinous nature pH being approximately 6. The technique of drying this product without any damage to the nature of it has not been perfected yet. But the alginic acid could be stored in a refrigerator in this fresh form for more than two months without any change. Further investigations have to be carried out to study the feasibility of commercialising these two products. Also experiments have to be carried out to cultivate these algae in larger quantities in order to make the production of agar and alginic acid more economical.

Acknowledgement.

Our sincere thanks are due to Mr. T. Kathirkamarajah for his technical assistance.

References

1. DURAIRATNAM, M. (1961) *Contribution to the study of the marine algae of Ceylon* – Bulletin Number **10**
2. RAO, A. V. & MODI, I. C. (1965) *Indian J. Technol*, **3**: 261 – 262.
3. RAO, A. V., PATEL K. N. & SHAH, H. N. (1965) *Research and Industry*, **10**: 131 – 133.
4. WOOD, J. F., (1946) *Agar in Australia* – Council for Scientific and Industrial Research Bulletin No. 203.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1)

Carbohydrate Constituents of the Marine Algae of Sri Lanka Part I. Some Physico - chemical properties of Phycocolloids from Eight Species of Red Algae

A. P. DANTANARAYANA, N. SAVITRI KUMAR AND M. U. S. SULTANBAWA

Department of Chemistry, University of Peradeniya, Peradeniya, Sri Lanka.

AND

S. BALASUBRAMANIUM

Department of Botany, University of Peradeniya, Peradeniya, Sri Lanka.

(Date of receipt : 11 March 1980)

(Date of acceptance : 03 November 1980)

Abstract: Phycocolloids from eight species of red algae were extracted, and the yields estimated. *Gracilaria edulis (lichenoides)* and *Gracilaria salicornia* were found to be the most suitable for commercial exploitation. The gel strength properties and their relationship to chemical constituents of the phycocolloids are discussed in relation to similar properties of a commercially available sample of agar.

1. Introduction

Agar and the alginates are two phycocolloids of commercial importance. Agar is extracted from red algae, whilst alginates are obtained from brown algae.

The polysaccharides, agarose and agaropectin are the main constituents of Agar. Agarose is a linear chain of alternating neutral sugar residues – mainly 1, 3 linked D–galactose and 1, 4 linked 3, 6– anhydrous L-galactose residues. Agaropectin consists mainly of D–galactose, 3,6-anhydro-L–galactose, some ester sulphate and D–glucuronic acid. It has many of the structural features of agarose.

Agar is insoluble in cold water but dissolves readily in boiling water, and sets to a firm gel at concentrations as low as 0.5%. It is a valuable colloidal substance because of its hydrophilic nature and its high gel strength. These two important properties are responsible for its wide use in the food, pharmaceutical and textile industries as a thickening, emulsifying, stabilizing and gelling agent. It is also used in medical and bacteriological laboratories as a culture medium for microorganisms.

More than 50 species of red algae have been used as raw materials in the manufacture of agar. Principal sources are species of *Gelidium*, *Gracilaria*, *Ahnfeltia*, *Pterocladia* and *Phyllophora*. In Japan, the largest amount of agar, is extracted mainly from plants of the genus *Gelidium*.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Durairatnam and Medcof[1] made a preliminary survey of the Sri Lankan sources of algae for the extraction of agar on an industrial scale. Durairatnam[2] after a systematic survey reported that *Gracilaria confervoides* and *Gracilaria edulis (lichenoides)* were found in large quantities, had a high gel content and could be used for the production of agar. We now report a preliminary study made of the feasibility of extracting phycocolloids from eight species of red algae collected in Sri Lanka and some physico-chemical properties of these phycocolloids.

2. Experimental Methods and Materials

The locality, habitat and distinguishing morphological features of the eight species of red algae studied are given in Table 1. Of the eight species studied, two species showed some biological variation. Brown and red thalli of *Gracilaria edulis*, and also orange and green thalli of *Hypnea musciformis* were collected. But for the extraction of phycocolloids these samples were bulked and replicate subsamples (50 g) were used. *Gracilaria corticata* is a polymorphic species but data reported in this paper refer to one form collected from Hikkaduwa.

The seaweed samples were washed and thoroughly sun dried (2-3 days) close to the site of collection. The problem of sample deterioration due to microbial activity was not observed. The dried samples were ground in a mill and extracted according to the procedure used by Durairatnam *et al.*[3] The sun dried product was stirred successively with ethanol and acetone.

The moisture content of the sun dried samples was determined by heating the samples at 110°C for 4 hours and the ash content obtained by incinerating the samples at 550°C for 5 hours. The water soluble Na^+, K^+, and Ca^{++} salts present in the ash of the algae and the agar, respectively, were recorded by using an EEL flame photometer.[6] The sulphate content was determined as $BaSO_4$ after a preliminary treatment with 0. 5 N HCl. The gel strength, temperature of gel formation and the melting temperature of the gel were determined as described by Ramarao and Krishnamoorthy.[6]

3. Results and Discussion

Table 2 gives the percentage yield and colour of the phycocolloids isolated. The best yield of phycocolloid was obtained from *Gracilaria salicornia*. All other species of *Gracilaria* studied gave reasonable yields of phycocolloids. Of these only G. *salicornia* and G. *edulis (lichenoides)* can be gathered or collected in large quantities and hence be considered worthwhile for commercial exploitation.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 1 – Locality, habitat and morphological features of red algae

Species	Locality	Habitat	Form and size of thallus
1. Gracilaria corticata	Hikkaduwa	Protected calcareous reef	10 cm, flat, dichotomous, fastigiate cartilaginous thallus
2. Gracilaria fergusonii	Hambantota	Exposed rocky boulders	ca. 7 cm, cylindrical, pseudodichotomous, cartilaginous
3. Gracilaria crassa	Koggala	Calcareous reef	ca. 8 cm, cylindrical, dichotomous, succulent, arched, decumbent
4. Gracilaria salicornia	Jaffna	Muddy lagoons	Grows in clumps, 20–30 cm wide, 10–15 cm high, greenish yellow, cylindrical with constrictions
5. Gracilaria edulis (lichenoides)	Jaffna	Protected inshore lagoons	10–30 cm, high, cylindrical, tapers to a point, irregularly dichotomous, distichous brown/green
6. Acanthophora delile	Jaffna	Lagoon	10–15 cm, cylindrical, covered with spiny outgrowths, drying black.
7. Gelidiella acerosa	Jaffna	Intertidal reef	5–9 cm, cylindrical, alternate, pinnate, wiry and cartilaginous
8. Hypnea musciformis	Jaffna	Protected inshore waters	ca. 8 cm, cylindrical, irregularly branched, tips of branches curled, membranous.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 2 – Moisture, ash and sulphate contents

Species	Yield % of crude phycocolloid	colour of phycocolloid	Moisture (%)		Ash (%)		Sulphate (%)	
			Algal material	Phyco-colloid	Algal material	Phyco-colloid	Algal material	Phyco-colloid
1. Gracilaria corticata ..	51·3	pale brown	14·86	18·40	15·61	6·77	2·71	2·93
2. Gracilaria fergusonii ..	53·0	pale brown	15·25	13·70	8·76	8·37	2·63	5·15
3 Gracilaria crassa ..	46·0	white	15·53	14·10	14·27	8·33	2·43	3·11
4. Gracilaria salicornia (cast ashore) ..	51·0	white	14·86	12·50	12·27	3·37	1·88	2·25
5. G. salicornia ..	62·0	white	13·43	16·80	14·54	4·04	1·92	2·43
6. Gracilaria edulis (lichenoides) ..	47·0	off white	13·73	18·50	15·90	5·05	4·23	2·27
7. Acanthophora delile ..	29·6	dark brown	16·38	21·50	12·97	14·00	5·55	10·25
8. Gelidiella acerosa ..	48·0	white	15·91	13·90	12·78	11·29	0·62	0·79
9. Hypnea musciformis ..	30·0	white	17·30	13·50	13·07	11·27	4·19	5·09
10. Spanish agar ..	—		—	7·50	—	1·69	—	0·64

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

The moisture, ash and sulphate contents of the algae and the phycocolloids isolated are recorded in Table 2. The sulphate content of the phycocolloids from *Gracilaria fergusonii*, *Acanthophora delile* and *Hypnea musciformis* was noticeably higher than that of the other species investigated, while that from *Gelidiella acerosa* was significantly low and comparable to that of Spanish agar.

Table 3 gives the percentage of the water soluble sodium, potassium and calcium ions present in the ash from the algal and phycocolloidal material respectively. It is interesting to note that the ash of the phycocolloid from G. *fergusonii* and G. *crassa* has a very high concentration of potassium while those of G. *corticata* and G. *edulis (lichenoides)* is relatively low.

Table 3 – Water soluble Na, K and Ca contents of the ash

Species	Algal material			Phycocolloid		
	% Na	% K	% Ca	% Na	% K	% Ca
1. Gracilaria corticata ..	7·50	26·50	3·20	3·20	0·72	4·32
2. Gracilaria fergusonii	8·50	11·00	2·65	3·57	8·50	2·50
3. Gracilaria crassa	9·00	11·00	2·80	3·60	8·50	1·85
4. Gracilaria salicornia (cast ashore) ..	12·50	27·00	3·10	1·80	2·00	1·80
5. G. salicornia ..	9·75	20·00	4·90	5·35	3·00	2·60
6. Gracilaria edulis (lichenoides) ..	7·80	19·25	2·55	2·70	1·50	0·92
7. Acanthophora delile ..	8·50	12·50	4·32	5·00	5·00	3·50
8. Gelidiela acerosa ..	15·00	26·00	4·10	3·10	2·60	4·60
9. Hypnea musciformis ..	17·00	4·50	5·85	4·50	2·50	1·95
10. Spanish agar ..	—	—	—	6·50	0·35	2·90

The gel strength, temperature of gel formation and the melting temperature of the gels formed by 1.0%, 1.5%, 2.0% and 2.5% solutions respectively, are given in Table 4. In general, species with high ash content also have a high sulphate content and correspondingly low gel strength, eg. *Acanthophora delile* and *Hypnea musciformis*. Further phycocolloids with a high content of water soluble potassium salts appear to form gels with better properties. Thus the phycocolloids from G. *fergusonii*, G. *crassa* and G. *salicornia* yield gels with a wider setting and melting temperature range. The low gel strength of the sample from G. *fergusonii* is probably due to its high sulphate content. The low potassium content of the phycocolloid from G. *corticata* may be responsible for its low gel strength. It may be possible to improve the setting properties of such a phycocolloid by the addition of a soluble potassium salt.[6]

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 4 – Gelling properties of the phycocolloids

Species	1·0% solution			1·5 % solution			2·0% solution			2·5% solution		
	T_1 (°C)	T_2 (°C)	gel strength ($1/cm_2$)	T_1 (°C)	T_2 (°C)	gel strength ($g/cm)_2$	T_1 (°C)	T_2 (°C)	gel strength (g/cm_2)	T_1 (°C)	T_2 (°C)	gel strength (g/cm_2)
1. Gracilaria corticata	27	38	* * *	31	43	3·72	36	46	7·69	39	48	13·52
2. Gracilaria fergusonii	37	63	5·70	41	65	10·85	43	68	22·35	45	69	36·20
3. Gracilaria crassa	37	55	16·21	43	60	64·14	44	63	139·85	46	65	281·77
4. Gracilaria salicornia (cast ashore)	36	47	3·10	40	52	12·86	44	56	43·64	46	60	59·07
5. G. salicornia (fresh)	35	46	6·10	43	55	53·57	45	60	76·24	47	67	148·87
6. Gracilaria edulis (lichenoides)	40	49	13·18	42	55	88·39	45	58	122·36	46	63	262·21
7. Acanthophora delile		No gel		No gel			30	53	6·27	36	55	12·53
8. Gelidiella acerosa	33	41	3·83	38	52	10·54	41	58	43·64	43	62	54·55
9. Hypnea musciformis	66	34	* * *	28	36	* * *	33	37	5·69	33	39	9·23
10. Spanish agar	40	60	132·00	41	64	290·51	43	69	378·66	45	73	596·29

T_1 — setting temperature of gel
T_2 — melting temperature of gel
* Could not be determined.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

The gel strength of a gel is determined largely by the amount of polymeric material which has precipitated in the form of a network. It has been reported that 3, 6-anhydrogalactose is less hydrophilic than galactose.[4] Therefore galactans having a high 3, 6-anhydrogalactose content yield very strong gels.[4] Furthermore, the presence of sulphate groups makes the galactans more soluble and thus lowers the gel strength and the melting temperature of the gel, while the presence of K^+ will lead to an increase in gel strength. This has been attributed to the fact that potassium salts of D-galactose sulphates are less hydrophilic than the sodium and barium salts of these compounds.[5]

A combination of these three factors together with the presence of inorganic impurities may be responsible for the variation in gelling properties observed during this investigation.

The potassium content of the Spanish agar sample is very low compared to that of the phycocolloid samples prepared in our laboratory. Hence the potassium content alone cannot markedly affect the gelling properties of these substances. The superior gelling properties of this sample is probably attributable to its 3, 6-anhydrogalactose content of the agarose fraction and low sulphate content. The absence of inorganic impurities which is reflected in the very low value for its ash content, is another factor which must be taken into account. Improvement of the gel forming ability and gel strength properties of the phycocolloids isolated is part of our future programme of research.

4. Conclusion

Of the red algae studied *Gracilaria edulis (lichenoides)* and G. *salicornia* appear to be the most suitable for commercial exploitation.

Acknowledgements

We thank the National Science Council of Sri Lanka and the International Foundation for Science for financial assistance.

References

1. DURAIRATNAM, M. & MEDCOF, J. C. (1954) *Ceylon Trade Journal* **19** (4) : 1
2. DURAIRATNAM, M., (1961) *I. P. F. C. Occasional Paper* **61**: 4.
3. DURAIRATNAM, M., GRERO, J. & WIMALSIRI, P. (1972) *Bull. Fish. Res. Stn. Sri Lanka (Ceylon)*, **23**: 29
4. HJERPEN, S. (1964), *Biochim. biophys. Acta*, **79**: 393.
5. PAINTER, T. J. (1966), *In Proc. Int. Seaweed symp. Halifax Nova Scotia* (F. G. Young and J. L. McLachlan ed.), p. 305, Pergamon Press, Oxford.
6. RAMARAO, K. & KRISHNAMOORTHY, V. (1968), *Botanica Marina* **11**: 129.
7. VOGIL, A. I. (1962) *Quantitative Inorganic Analysis*, 3rd edition, p. 882–885. ELBS and Longmans, London.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1):

Convergence Factors in an Integral Mean

C. YOGACHANDRAN

Department of Mathematics, University of Peradeniya, Peradeniya, Sri Lanka.

(Date of receipt : 15 October 1979)

(Date of acceptance : 17 November 1980)

Abstract : The main theorem proved in this paper gives conditions necessary and sufficient in order that, for some real C_m $(m = 0, 1, \cdots r-1)$

$I_r f(x)\, g(x) = \sum_{m=0}^{r-1} C_m x^{r-1-m} + o(x^{p+q+r})$ whenever

$I_k f(x) = \sum_{m=0}^{k-1} c_m x^{k-1-m} + o(x^{p+k})$, where $I_k f(x)$ denotes the k-th iterated integral of $f(x)$, under suitable restrictions on the function f, g, and under the assumptions

$r, k \in N_0 \quad r \geq k \geq 0$, $p, q \in R$ and $p + q \notin \{-1-2, \cdots -r\}$.

1. Introduction

If $f \in L_{loc}$, $k \in N$, let $I_k f(x) = f_k(x)$ denote th k - the iterated integral of $f(x)$. We assume in this paper that $r, k \in N_0$, $r \geq k$, $p, q \in R$ and that (a) if $r = k = 0$ then $f, g \in F$ (The space of all real functions defined on $[1, \infty)$).

(b) if $k = 0$, $r \geq 1$, then $f \in L_{loc}$, $g \in B_{loc}$ (Space of functions in F bounded and measurable locally),

(c) if $r \geq k \geq 1$, then $f \in L_{loc}$, $g^{k-1} \in AC_{loc}$ (Space of functions in F absolutely continuous locally),

g^m denoting the m - th derivative of g, g^0 denoting g.

We prove the following theorems; omitting the trivial case $r = k = 0$.

Theorem 1. Let $p + q > -1$.

(a)If $k = 0$, $r \geq 1$,a necessary and sufficient condition that $I_r f(x)\, g(x) = 0(x^{p+q+r})$ whenever $I_k f(x) = 0(x^{p+k})$ is that

$$(i)_0 \quad 0 \int_1^x t^{-q} |g(t)|\, dt = 0(x) \text{ as } x \to \infty.$$

(b) If $r \geq k \geq 1$, then necessary and sufficient conditions that $I_r f(x)\, g(x) = 0(x^{p+q+r})$ whenever $I_k f(x) = 0(x^{p+k})$ are:

$$(i) \quad g(x) = 0(x^q) \text{ as } x \to \infty,$$

$$(ii) \quad \int_1^x t^{k-q} |g^k(t)| dt = 0(x) \text{ as } x \to \infty.$$

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

We find that the conditions in Theorem 1 are necessary even in the case $p + q \leqslant -1$. We also find that if $p + q < -1$, $p + q \notin \{-1, -2, \cdots -r\}$ and the conditions of Theorem 1 hold, then there exist constants C_m, $m = 0, 1, \cdots r-1$ such that $I_r f(x)\, g(x) = \sum_{m=0}^{r-1} C_m x^{r-1-m} + 0(x^{p+q+r})$.

We combine these results to give :

Theorem 2

(a) If $r \geqslant k \geqslant 0$, $r \geqslant 1$, $p + q \notin \{-1, -2, \cdots -r\}$, c_m $(m = 0, 1, \cdots k-1)$ are real, $I_k f(x) = \sum_{m=0}^{k-1} c_m x^{k-1-m} + 0(x^{p+k})$, and the conditions of Theorem 1 hold, then there exist constants C_m $(m = 0, 1, \cdots r-1)$ such that

$$I_r f(x)\, g(x) = \sum_{m}^{r-1} c_m x^{r-1-m} + 0(x^{p+q+r}).$$

(b) If $r \geqslant k \geqslant 0$, $r \geqslant 1$, and there exist constants C_m $(m = 0, 1, \cdots r-1)$ such that $I_r f(x)\, g(x) = \sum_{m=0}^{r-1} C_m x^{r-1-m} + 0(x^{p+q+r})$ whenever $I_k f(x) = \sum_{m=0}^{k-1} c_m x^{k-1-m} + 0(x^{p+k})$, where c_m $(m = 0, 1, \cdots k-1)$ are real, then the conditions of Theorem 1 are necessary. An empty sum denotes zero.

Theorem 2 is the analogue for continuous variables of the sequence – sequence problem considered in [1].

2. Auxiliary Results

Lemma 1. Let $k \in N$, $q \in R$ and $g^{k-1} \in AC_{loc}$. Then, each of the following sets of conditions :

(a) $(i)_a \int_1^x t^{-q} |g(t)|\, dt = 0(x)$ $\quad$ $(ii)_a \int_1^x t^{k-q} |g^k(t)|\, dt = 0(x)$,

(b) $(i)_b \int_1^x t^{-q} g(t)\, dt = 0(x)$ $\quad$ $(ii)_b \int_1^x t^{k-q} g^k(t)\, dt = 0(x)$.

(c) $(i)_c\ g(x) = 0(x^q)$ $\quad$ $(ii)_c\ g^{k-1}(x) = 0(x^{q+1-k})$,

implies that $g^j(x) = 0(x^{q-j})$ for $j = 0, 1 \cdots k-1$.

Proof. Clearly, it is enough to prove that $(i)_b$ and $(ii)_b$ imply that

$$g^j(x) = 0(x^{q-j}).$$

Case 1. Let $q > k-1$.

Since $g^{k-1}(x) = g^{k-1}(1) + \int_1^x t^{q-k} [t^{k-q} g^k(t)]\, dt$, partial integration and $(ii)_b$ give $g^{k-1}(x) = 0(x^{q-k+1})$, and further integration gives the result.

Case 2. $q = k-1$. Integration $(ii)_b$ $n-1$ times where $1 \leqslant n \leqslant k$, we get

$$I_n [x g^k(x)] = 0(x)^n \qquad (2 \cdot 1)$$

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Now, $(d/dx)^u\,[xg^{k-n}(x)] = xg^k(x) + ng^{k-1}(x)$ p. p., and integrating this n times we have

$$xg^{k-n}(x) - nI\,[g^{k-n}(x)] = 0(x^n), \tag{2·2}$$

by (2·1).

Now, $(i)_b$ and partial integration give $I\,[g(x)] = 0(x^k)$. (2·3)

Putting $n = k$ in (2·2) and using (2·3), we get

$$g(x) = 0(x^{k-1}). \tag{2·4}$$

Putting $n = k-1$ in (2·2) and using (2·4) we get $g^1(x) = 0(x^{k-2})$, and proceeding similarly we establish the result.

Case 3. $q < k-1$. Partial integration and $(ii)_b$ give, in this case,

$$g^{k-1}(x) = c + 0(x^{q-k+1}) = c + 0(1).$$

Hence $$\int_1^x t^{-q} g(t)\,dt = \frac{cx^{k-q}}{(k-q)(k-1)!} + 0(x^{k-q}),$$

and thus, $(i)_b$ can hold only if $c = 0$, and (2·5) gives

$g^{k-1}(x) = 0(x^{q-k+1})$, which is the conclusion if $k = 1$.

If $k \geqslant 2$, since $\int_1^x t^{k-1-q} g^{k-1}(t)\,dt = 0(x)$, in the case $q \geqslant k-2$, we can use cases 1 and 2 with k replaced by $k-1$ to get

$g^j(x) = 0(x^{q-j})$ for $j = 0, 1, \cdots k-2$. If $q < k-2$, by repetition of the above argument, we get $g^{k-2}(x) = 0(x^{q-k+2})$, and thus

$\int_1^x t^{k-2-q} g^{k-2}(t)\,dt = 0(x)$. We now establish the result by an inductive argument.

Lemma 2. If $d_{ij} = (-1)^j \binom{k}{j} \frac{u^{r-1+i-j}}{(r-1+i-j)!}$, $i, j = 0, 1, \cdots k$, $r \in N$,

and D_{ij} is the cofactor of d_{ij} in the determinant $D = \det(d_{ij})$ of order $(k+1) \times (k+1)$, then $D_{ij} = K_{ij}\,u^{k(r-1)+j-i}$, where K_{ij} is independent of u, $D = Pu^{(k+1)(r-1)}$, $P = \prod_{m=0}^{k} \frac{(-1)^m \binom{R}{m} m!}{(r-1+m)!} \neq 0$.

This result is proved easily by induction with respect to k.

Lemma 3. If $p + k > -1$, $k^1 > k$ and $g^k(x) = 0(x^{p+k})$, then

$$g_{k1}(x) = o(x^{p+k1}).$$ 'o' may be replaced by '0' here

This result is well known.

Lemma 4. If $p + k > -1$, $p + q > -1$ and $g_k(x) = o(x^{p+k})$, then $I_k x^q g(x) = o(x^{p+q+k})$. 'o' may be replaced by '0' here.
of [2] (Lemma 1) and [3] (Lemma 4).

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Lemma 5. Let $n \in N$ and let S_0^n be the subset of F such that every $s \in S_0^n$ satisfies $s^n \in AC_{loc}$ and $s(t) = o(1)$ as $t \to \infty$. Suppose $a(x, t) = 0$ for $t > x$, $a(x, t) \in L[1, x]$ for every fixed $x \geq 1$, and $\int_1^T |a(x, t)|\, dt = o(1)$ as $x \to \infty$ for every $T > 1$. Then, in order that $v(x) = \int_1^x a(x, t)\, s(t)\, dt = o(1)$ as $x \to \infty$ whenever $s \in S_0^n$, $s(1) = s^1(1) = \cdots = s^n(1) = o$ it is necessary that

$$\int_1^x |a(x, t)|\, dt = 0(1) \text{ as } x \to \infty.$$

See [4] (Lemma 8).

Lemma 6. Suppose $a(x, t)$ is bounded and measur able on $[1, x]$ for every $x > 1$ and $a(x, t) = 0$ for $t > x$. Then in order that $v(x) = \int_1^x a(x, t)\, s(t)\, dt = o(1)$ as $x \to \infty$ whenever $s \in L$ and $s(t) = o(1)$ as $t \to \infty$, it is necessary that $\int_1^x |a(x, t)|\, dt = 0(1)$ as $x \to \infty$.

3. Proofs of Theorems

Proof of Theorem 1. Necessity.

By repeated partial integration, when $r \geq k \geq 1$, we have

$$I_r f(x)\, g(x) = \int_1^x \frac{(x-t)^{r-1}}{(r-1)!} f(t)\, g(t)\, dt$$

$$= (-1)^k \int_1^x f_k(t)\, (D_t)^k G_r(x, t)\, dt + \delta_{rk} f_k(x)\, g(x), \qquad (3\cdot1)$$

where $D_t \equiv \frac{\partial}{\partial t}$, $G_r(x, t) = \frac{(x-t)^{r-1}}{(r-1)!} g(t)$, and δ_{ik} is the Krönecker delta.

When $k = 0$, $r \geq 1$, the formula still holds.

Define $a(x, t) = (-1)^k t^{p+k} x^{-p-q-r} (D_t)^k G_r(x, t)$, $s(t) = t^{-p-k} f_k(t)$

and $v(x) = \int_1^x a(x, t)\, s(t)\, dt$.

Then, (3·1) gives

$$x^{-p-q-r} I_r f(x)\, g(x) = v(x) + \delta_{rk}\, x^{-p-q-r} f_k(x)\, g(x). \qquad (3\cdot2)$$

Since conditions $(i)_0$, (i) and (ii) are independent of r, $r \geq \max(1, k)$, it is enough, by Lemma 3, to consider the case $r \geq k + 1$ here, and then (3·2) gives:

$v(x) = o(1)$ as $x \to \infty$ whenever $s \in S_0^{k-1}$ when $k \geq 1$, and

$v(x) = o(1)$ whenever $s \in L$ and $s(t) = o(1)$, when $k = 0$, where S_0^{k-} is defined in Lemma 5.

Clearly, $a(x, t)$ satisfies the conditions of Lemma 5 and 6, and hence we get the necessary condition $\int_1^x |a(x, t)|\, dt = 0(1)$, which gives $\int_1^x t^{p+k} |(D_t)^k G_r(x, t)|\, dt = 0(x^{r+q+r})$, $k \geq 0$, and by Lemma 3, r may be replaced by $r + 1$, $i = 0, 1, \cdots$ in this equation.

Thus, $$\int_1^x t^{p+k} |D_t)^k G_{r+i}(x, t)|\, dt = 0(x^{p+q+r+i}). \qquad (3.3)$$

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Now $(D_t)^k G_{r+i}(x,t) = \sum_{j=0}^{k} \frac{(-1)^j \binom{k}{j} (x-t)^{r+i-j-1}}{(r+i-j-1)!} g^{k-j}(t)$, $i = 0, 1, \cdots k$,

and solving these equations and using Lemma 2, we get

$$P(x-t)^{(k+1)(r-1)} g^{k-j}(t) = \sum_{i=0}^{k} D_{ij} (D_t)^k G_{r+i}(x,t)$$

$$= \sum_{i=0}^{k} K_{ij} (x-t)^{k(r-1)+j-i} (D^t)^k G_{r+i}(x,t), \text{ for } j = 0, 1, \cdots k, \text{ where } K_{ij}$$

is independent of x and t, $P \neq 0$.

Hence, since $k(r-1) + j - i \geqslant 0$, we have

$$(\tfrac{1}{2}x)^{(k+1)(r-1)} \int_1^{x/2} t^{k+p} |g^{k-j}(t)| dt$$
$$\leqslant \int_1^{x/2} t^{k+p} (x-t)^{(k+1)(r-1)} |g^{k-j}(t)| dt$$
$$< \int_1^x t^{k+p} (x-t)^{(k+1)(r-1)} |g^{k-j}(t)| dt$$
$$\leqslant \sum_{i=0}^{k} \left|\frac{Kij}{P}\right| x^{k(r-1)+j-i} \int_1^x t^{p+k} |(D_t^k) G_{r+i}(x,t)| dt$$
$$= 0(x^{(k+1)(r-1)+p+q+1+j}) \text{ for } j = o, 1, \cdots k, \text{ by } (3\cdot3), (4\cdot4)$$

Now, $(3\cdot4)$ gives

$\int_1^x t^{k+p} |g^{k-j}(t)| dt = 0(x^{p+q+1+j})$, $j = 0, 1, \cdots k$, and by Lemma 4,

this gives $\int_1^x t^{k-q-j} |g^{k-j}(t)| dt = 0(x)$. $(3\cdot5)$.

If $k \geqslant 1$, taking $j = 0$ and $j = k$ in $(3\cdot5)$ and applying Lemma 1, we get the necessary conditions (i) and (ii).

If $k = 0$, we get the single condition $(i)_0$ from $(3\cdot5)$.

Sufficiency. By expanding $(x-t)^{r-1}$ and applying Leibniz's theorem,

we get $(-1)^k (D_t)^k G_r(x,t) = \sum_{m=0}^{r-1} A_m x^{r-1-m} \sum_{i=0}^{k} B_i t^{m-i} g^{k-i}(t)$ when $k \geqslant 1$

and $(-1)^k (D_t)^k G_r(x,t) = \sum_{m=0}^{r-1} A_m x^{r-1-m} t^m g(t)$ when $k = 0$, where

$$A_m = \frac{(-1)^{m+k}\binom{r-1}{m}}{(r-1)!}, \quad B_i = \binom{k}{i} m(m-1)\cdots(m-i+1) \text{ for } i \geqslant 1, B_0 = 0.$$

Substituting in $(3\cdot1)$, we get, when $k \geqslant 1$, $I_r f(x) g(x)$

$$= \sum_{m=0}^{r-1} A_m x^{r-1-m} \sum_{i=0}^{k} B_i \int_1^x t^{m-i} f_k(t) g^{k-i}(t) dt + \delta_{rk} f_k(x) g(x), \qquad (3\cdot6)$$

and $I_r f(x) g(x) = \sum_{m=0}^{r-1} A_m x^{r-1-m} \int_1^x t^m f(t) g(t) dt$ when $k = 0$. $(3\cdot7)$

If $k = 0$ and $(i)_0$ holds, Lemma 4 gives

$\int_1^x t^{p+m} |g(t)| dt = 0(x^{p+q+m+1})$ for $m = 0, 1, \cdots r-1$, and since $f(t) = o(t^p)$,

$\int_1^x t^m f(t) g(t) dt = \int_1^x o(t^{p+m}) |g(t)| dt = 0(x^{p+q+m+1})$, and hence

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

(3·7) gives $I_r f(x) g(x) = o(x^{p+q+r})$, the required result.

If $k \geq 1$, (i) and (ii) hold, by Lemma 1 we get

$\int_1^x t^{p+k+m-i} |g^{k-i}(t)| dt = 0(x^{p+q+m+1})$, and since $f_k(x) = o(x^{p+k})$,

$\int_1^x t^{m-i} f_k(t) g^{k-i}(t) dt = o(x^{p+q+m+1})$, for $i = 0, 1, \cdots k$, and

substituting in (3·6) we get

$$I_r f(x) g(x) = o(x^{p+q+r}) + o(x^{p+q+k}) = o(x^{p+q+r}).$$

Proof of Theorem 2.

(a) We may take, without loss of generality, $c_m = 0$ for $m = 0, 1, \cdots k-1$,

since $f(x)$ may be replaced by $h(x) = f(x) - \sum_{m=0}^{k-1} c_m^1 e^{-(m+1)x}$,

so that $h_k(x) = f_k(x) - \sum_{m=0}^{k-1} c_m x^{k-1-m} = o(x^{p+k})$, (the constants c_m^1 being chosen suitably), while the form of the conclusion is unchanged.

It is also enough to take $p + q < -1$, $p + q \notin \{-1, -2, \cdots -r\}$.

If $k = 0$ and $(i)_0$ holds, as in the sufficiency part of Theorem 1,

we get $\int_1^x t^m f(t) g(t) dt = o(x^{p+q+m+1}) + a_m$ for $m = 0, 1, \cdots r-1$, and

substituting in (3·7) we get

$$I_r f(x) g(x) = \sum_{m=0}^{r-1} C_m x^{r-1-m} + o(x^{p+q+r}), \text{ where } C_m = A_m a_m.$$

If $k \geq 1$ and (i) and (ii) hold, as before we have

$\int_1^x t^{m-i} f_k(t) g^{k-i}(t) dt = o(x^{p+q+m+1}) + a_{mi}$, $i = 0, 1, \cdots k$ and

$m = 0, 1, \cdots r-1$. Substituting in (3·6) we get the conclusion

with $C_m = A_m \sum_{i=0}^{k} B_i a_{mi}$. Clearly, every $C_m = 0$ when $p + q > -1$.

(b) We may take every $c_m = 0$, $p + q \leq -1$. Let n be a positive integer such that $p + q + n > -1$.

By (a) above, with $f(x)$, $g(x)$, p, q, k replaced by $f(x)g(x)$, x^n, $p + q$, n, r respectively, we see that $I_r f(x) g(x) = \sum_{m=0}^{r-1} C_m x^{r-1-m} + o(x^{p+q+r})$

implies $I_r x^n f(x) g(x) = o(x^{(p+q)+n+r})$, since $(p + q) + n > -1$.

i. e. $I_r f(x) \cdot x^n g(x) = o(x^{p+(q+n)+r})$ whenever $f_k(x) = o(x^{p+k})$, and hence by Theorem 1, with $g(x)$, q replaced by $x^n g(x)$, $q + n$, we get the necessary conditions:

$$\int_1^x t^{-(q+n)} |t^n g(t)| dt = 0 \ (x) \text{ when } k = 0 \qquad (3\cdot8)$$

and

$$x^{-(q+n)} x^n g(x) = 0(1), \qquad (3\cdot9)$$

$$\int_1^x t^{k-(q+n)} |(d/dt)^k [t^n g(t)]| dt = 0(x) \text{ when } k \geq 1 \qquad (3\cdot10)$$

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

(3·8) and (3·9) are the same as $(i)_0$ and (i) respectively

By Lemma 1, (3·9) and (3·10) imply, for $j = 0, 1, \cdots k$,

$$\int_1^x t^{j-q-n} \left| (d/dt)^j [t^n g(t)] \right| dt = 0(x). \qquad (3\cdot11)$$

But, $$g^k(t) = (d/dt)^k [t^{-n} . t^n g(t)] = \sum_{j=0}^{k} d_j t^{-n-k+j} (d/dt)^j [t^n g(t)].$$

where $$d_j = \binom{k}{j} (-n)(-n-1)\cdots(-n-k+j+1).$$

Hence $$\int_1^x t^{k-q} |g^k(t)| dt \leqslant \sum_{j=0}^{k} |d_j| \int_1^x t^{-q-n+j} \left| \left(\frac{d}{dt}\right)^j [t^n g(t)] \right| dt$$

$$= 0(x), \text{ by } (3\cdot11).$$

This completes the proof.

References

1. BOSANQUET, L.S. (1954) *Mathematika* **1** ; 24—44.
2. BOSANQUET, L.S. (1948) *J. Lond. Math. Soc.* **23** ; 35—38.
3. BORWEIN, D. (1950) *J. Lond. Math Soc.* **25** ; 289—302.
4. BORWEIN, D. (1954) *J. Lond. Math. Soc.* **29** ; 276 – 292.
5. HARDY, G.H. (1949) 'Divergent Series' (Oxford U.P.)

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1) :

An Electron Microscopic Study of the Liver in the *Rhacophorus leucomystax maculatus (Gray)* Tadpole after Treatment with Aflatoxin B 1

A. D. P. JAYATILAKA AND S. KIRUPANANTHAN

Department of Anatomy, Faculty of Medicine, University of Peradeniya, Peradeniya, Sri Lanka.

(Date of receipt : 01 February 1980)

(Date of acceptance : 12 December 1980)

Abstract: Tadpoles, being visible to the naked eye, were chosen to observe embryonic material. *Rhacophorus leucomystax maculatus* tadpoles were treated with LD 50 of aflatoxin B1. Tadpoles were moribund on the 2nd, 3rd and 4th days after treatment. Those that survived the treatment were transfered into fresh distilled water. The livers of control tadpoles and those that were moribund on the 2nd, 3rd and 4th days were dissected by microdissection, processed and observed with an electron microscope. The livers of those tadpoles that survived the treatment were also processed after two weeks. The hepatocytes of the treated tadpoles showed nucleolar capping and in the cytoplasm there was fatty infiltration, structural changes in the organelles, presence of crystals and complete absence of glycogen; glycogen was present in the hepatocytes of control tadpoles and also in those tadpoles that survived treatment. The structural changes in the hepatocytes of treated tadpoles were probably due to the toxicity of aflatoxin on the cell.

1. Introduction

Several workers have observed the ultrastructural changes in the livers of various animals due to toxicity of aflatoxin B1. These observations have been made in the rat, [3,4,5,9] in the rat and in the monkey[8] and in the duckling. [10,11]

Arsecularatne *et al*[2] used tadpoles of the *Rhacophorus leucomystax maculatus* for the bioassay of aflatoxin B1. This was the first time that observations were made on the toxic effects of aflatoxin B1 on visually observable embryonic material when compared to other animals whose embryos were either in uteri or in eggs. Jayatilaka and Maxwell[6] observed electron microscopic changes in livers of *Xenopus laevis* (Daudin) tadpoles after treatment with aflatoxin B1. This paper presents the ultrastructural changes, caused by LD 50 of aflatoxin, in the liver of the *Rhacophorus leucomystax maculatus* tadpoles, in the same larval stages, as those used by Arsecularatne *et al*[2] in their study of the liver by light microscopy.

2. Materials and Methods

Tadpoles from a single spawn nest were reared and measured as described by Joseph and Jayatilaka.[7] 20 mm tadpoles (total length) were treated with 1·6 μg/ml (LD 50) aflatoxin B1 for 4 days as described by Arsecularatne *et al*.[2] Death

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

of tadpoles commenced about the 2nd day and was most frequent on the 3rd and 4th days. The livers were dissected by microdissection in control tadpoles and in moribund tadpoles on the 2nd, 3rd and 4th days. Tadpoles that survived were transferred to fresh distilled water on the 5th day and fed with spinach leaves thereafter. After a period of two weeks, the livers of these tadpoles were also dissected by microdissection. The dissected livers were immersed in a mixture containing 4% paraformaldehyde, 0·5% gluteraldehyde, 0·01% calcium chloride in 0·1 M sodium cacodylate at a pH of 7·3 for 24 hours at a temperature of 4°C. They were then postosmicated in a mixture containing 1·0% osmium tetroxide, 0·1% calcium chloride in 0·75 M sodium cacodylate at a pH of 7·37 for 2 hours at a temperature of 4° C. The specimens were then dehydrated in graded alcohols and finally embedded in epon. Ultra thin sections were cut on a Porter-Blum Ultramicrotome, floated on to copper grids, doubly stained with uranyl acetate and lead citrate mixture and examined under a Hitachi 12 U electron microscope operated at 75 KV.

3. Results

In the control tadpole, the hepatocytes showed a rounded nucleus with a nucleolus (Figure 1). Organelles such as mitochondria, rough endoplasmic reticulum, ribosomes, vacuoles and glycogen were observed in the cytoplasm (Figure 1). A large number of microvesicles were also observed in the cytoplasm (Figure 2). Tight junctional complexes were seen between adjoining hepatocytes which formed the bile canaliculi. Microvilli were seen to project into the lumen of a bile canaliculus (Figure 1).

The hepatocytes of larvae who were moribund on the 2nd day showed nucleolar changes and fatty infiltration of the cytoplasm (Figure 2). The nucleolus showed nucleolar capping with nucleolar material arranged in a peripheral very dense and less dense zones. The very dense zone was thicker than the less dense zone and the entire nucleolus gave the appearance of a signet ring (Figure 2). The centre of the nucleolus showed a clear area with fine granular material and extremely electron dense material in the middle of the clear area (Figure 2). A similar area was seen in the very dense part of the peripheral nucleolar material with fine granular material within it (Figure 2). The fat infiltration was seen as large ovoid cavities which were lined with electron dense material (Figure 2). The centre of the fat vacuole was clear. Macrovesicles were also observed in the cytoplasm. The hepatocytes of the tadpoles who were moribund on the 3rd day showed irregularly shaped nuclei and in the cytoplasm there were more fatty vacuoles either completely filled or

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

partly filled with electron dense material (Figure 3). Other hepatocytes showed fatty infiltration where the vacuoles were filled with dark electron dense material (Figure 3). The mitochondria in these cells were swollen and contained dark electron dense material (Figure 3). The rough endoplasmic reticulum and ribosomes were not observed but multivesicular and autophagic vacuoles were beginning to appear. The appearence of crystals bounded by membrane was another feature (Figure 3). The hepatocytes in tadpoles who were moribund on the 4th day were the seat of marked cellular degeneration. They also showed markedly distorted nuclei and the cytoplasm showed the presence of dilated vacuoles, some empty and others with membrane material, while autophagic vacuoles were distinctly more than in the hepatocytes of those tadpoles that were moribund on the 3rd day (Figure 4). In some hepatocytes of tadpoles, who were moribund on the 4th day, the cytoplasm showed dilated rough endoplasmic reticulum and were circular in cross section (Figure 5). These were identified as R. E. R. due to the presence of ribosomes attached to their membranes (Figure 5). Some hepatocytes showed densely osmophilic chromatin, in the nucleus and the cytoplasm showed the presence of fatty infiltration, swollen mitocondria, dilated smooth endoplasmic reticulum and multivesicular bodies. Other hepatocytes showed only the presence of multivesicular and autophagic vacuoles, fat globules, myelin figures and membrane bound vesicles (Figure 6). In other hepatocytes, the nucleus was prominent with a well marked condensed nucleolus. In these the cytoplasmic organelles were not readily recognisable except for fat globules, distended vacuoles and myelin figures (Figure 7). Another marked feature was the presence of a large number of microvilli projecting into the lumen of each bile canaliculus when compared to a bile canaliculus in a control tadpole (Figures 1 and 8). Some bile canaliculi were grossly distended with few microvilli projecting into the lumen. In these hepatocytes the nucleolus was swollen but did not show nucleolar capping. Glycogen was not present in the hepatocytes of those tadpoles moribund on the 2nd, 3rd and 4th days after treatment.

In those tadpoles that survived the treatment and who were sacrificed after two weeks, the hepatocyte showed completely normal cellular architecture and was similar to the hepatocytes of the control tadpoles. The nucleolus appeared normal and the cytoplasm showed the presence of all other organelles and the presence of glycogen.

4. Discussion

The electron microscopic findings in this study support the observations of Arsecularatne *et al*[2] by light microscopy. Nuclear changes and cytoplasmic vacuolation was very well marked in the present study. The hepatocytes showed various stages of cellular degeneration. The vesicular nuclei observed by Arsecularatne *et al*[2] was probably due to the dispersal of chromatin which showed as vacuolation in light microscopy.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

The findings in this study closely resembled the observation of Bassir and Babamunmi,[3] Butler [4, 5] and Svoboda and Higginson[9] in the rat, in the rat and in the monkey by Svoboda, Grady and Higginson[8] and in the duckling by Theron[10] and Theron, Liebenberg and Joubert.[11] Similar changes were observed by Jayatilaka and Maxwell[6] in the hepatocytes of *Xenopus laevis* (Daudin) tadpole. Nucleolar capping, dilatation of rough endoplasmic and smooth endoplasmic reticula, mitochondrial enlargement, multivesicular and autophagic vacuoles and bile canaliculi hyperplasia have also been observed in this study.

In eukaryocyte organisms, a definite number of chromosomes of the cell take part in organising the nucleolus–called nucleolar organisers. It is seen as perinucleolar chromatin. For this purpose, the chromosomes were located close to the nucleolus. Allison and Paton[1] have shown that enzymes liberated from lysosomes may enter the nucleus and produce chromosomal aberrations. These two workers have shown that lysosomal enzymes produce chromatid breaks and play a part in carcinogenesis. Lysosomal enzymes could gain access to genetic material and cause changes without impairing mitosis. Lysosomes can be affected by carcinogenic agents like aflatoxin B1. The granular element have been shown to be RNA and the fibrillar elements the DNA strands. Thus nucleolar capping may be the earliest changes seen in the nucleolus of those cells that may become cancerous due to a changed genetic structure.

The other observations were mainly due to the acute toxicity of the hepatocytes. The second day tadpoles showed fatty infiltration in the cytoplasm which was a characteristic feature in the hepatocytes of those tadpoles who were moribund on the 3rd and 4th days. The hepatocytes of the tadpoles who were moribund on the 2nd, 3rd and 4th day after treatment showed alteration of the structure of the organelles in various degrees, while in some hepatocytes the picture was that of cell death.

The hepatocytes of those tadpoles who survived the treatment showed normal structure This perhaps was due to the fact that the hepatocytes either had undergone regeneration in the liver or that these hepatocytes were not affected at all. Arsecularatne *et al*[2] reported that those tadpoles which survived became giant tadpoles and that the period of their metamorphoses was delayed. Thus it may be postulated that the metabolism of the tadpole was in some way deranged by the toxicity of the hepatocytes by aflatoxin B1 and this in turn may have caused disruption of the hypothalamo - hypophysio - thyroid axis thereby producing giant tadpoles and also delay in metamorphosis.

Acknowledgements.

We wish to thank Mr. R. M. C. Ekanayake, Electron Microscope Technician of the Electron Microscope Unit of the Faculty of Medical, Dental and Veterinary Sciences for his technical assistance and the National Science Council for providing funds to carry out this study.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

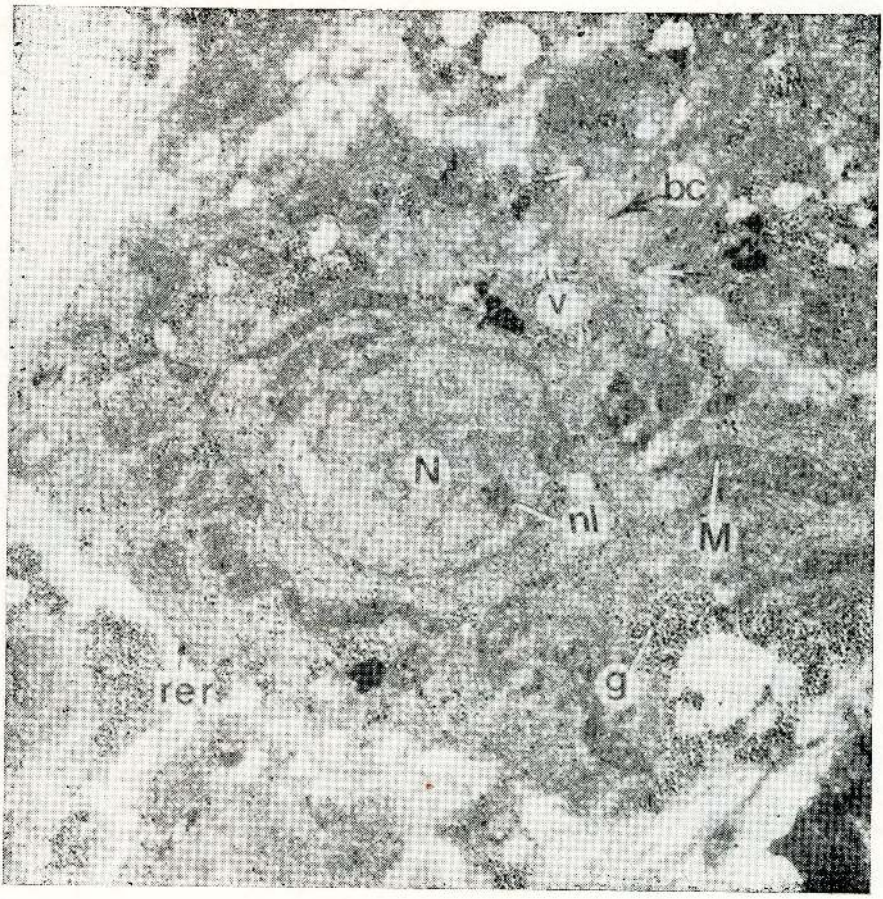
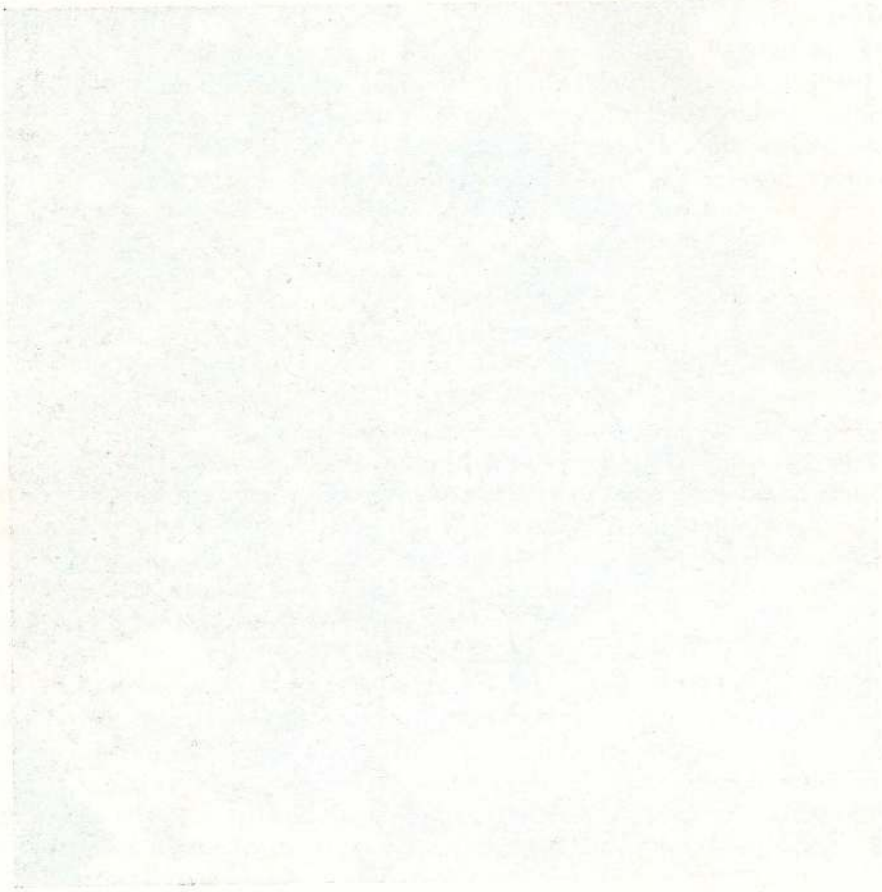


Figure 1. An electron micrograph of a hepatocyte in a control tadpole showing nucleus (N), nucleolus (nl), mitochondria (M), rough endoplasmic reticulum (rer), vacuole (V), and glycogen (g). Arrows show tight junctions near bile canaliculus (bc). x 10,250

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media



Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

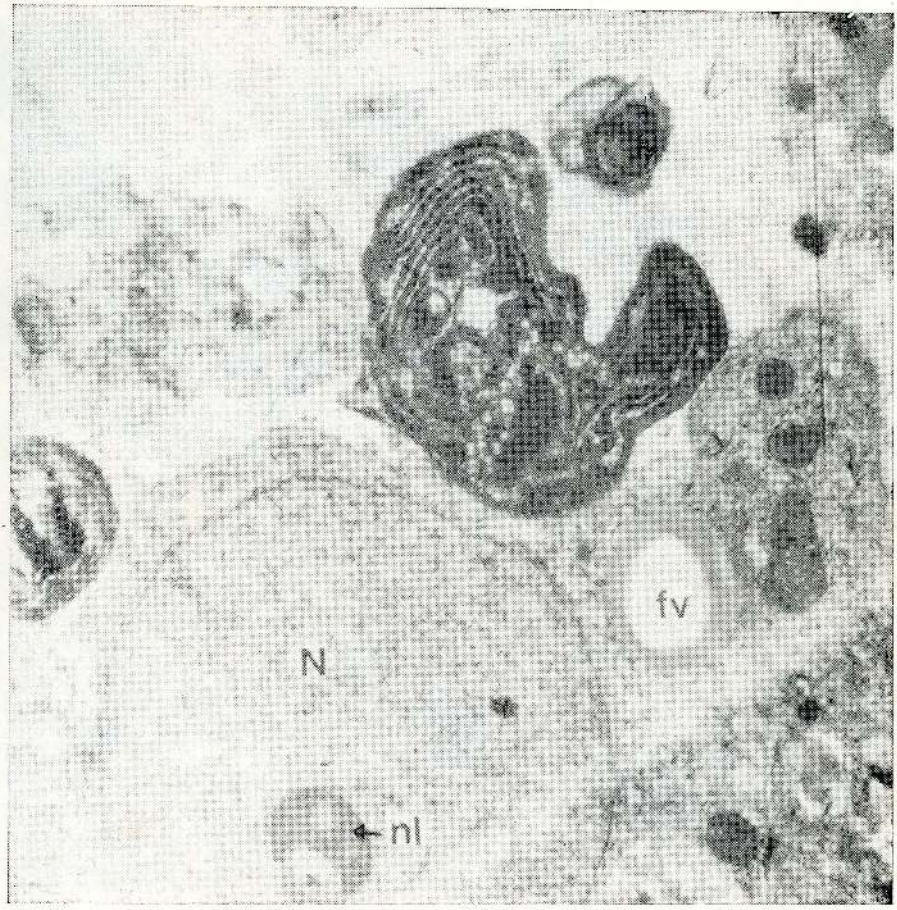


Figure 2. An electron micrograph of a hepatocyte in a tadpole moribund after two days of treatment showing nucleolar capping (nl) and a fat vacuole (fv) . x 16,000

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

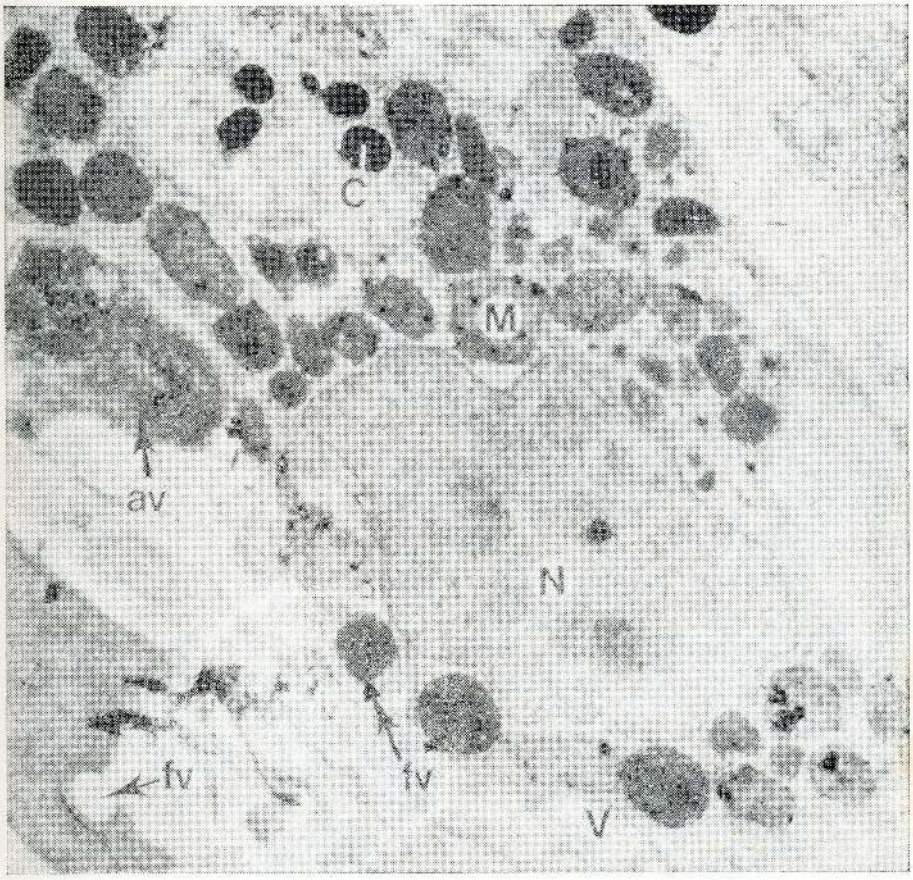


Figure 3. An electron micrograph of a hepatocyte in a tadpole moribund after three days of treatment showing irregularly shaped nucleus (N), swollen mitochondria (M), electron fat vacuoles (fv), vacuoles (V) autophagic vacuoles (av) and membrane bound crystals (C). x 16,000

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Figure 4. An electron micrograph of a hepatocyte in a tadpole moribund after four days of treatment showing distorted nuclei (N), dilated vacuoles (V) and autophagic vacuoles (av). x 12,750

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

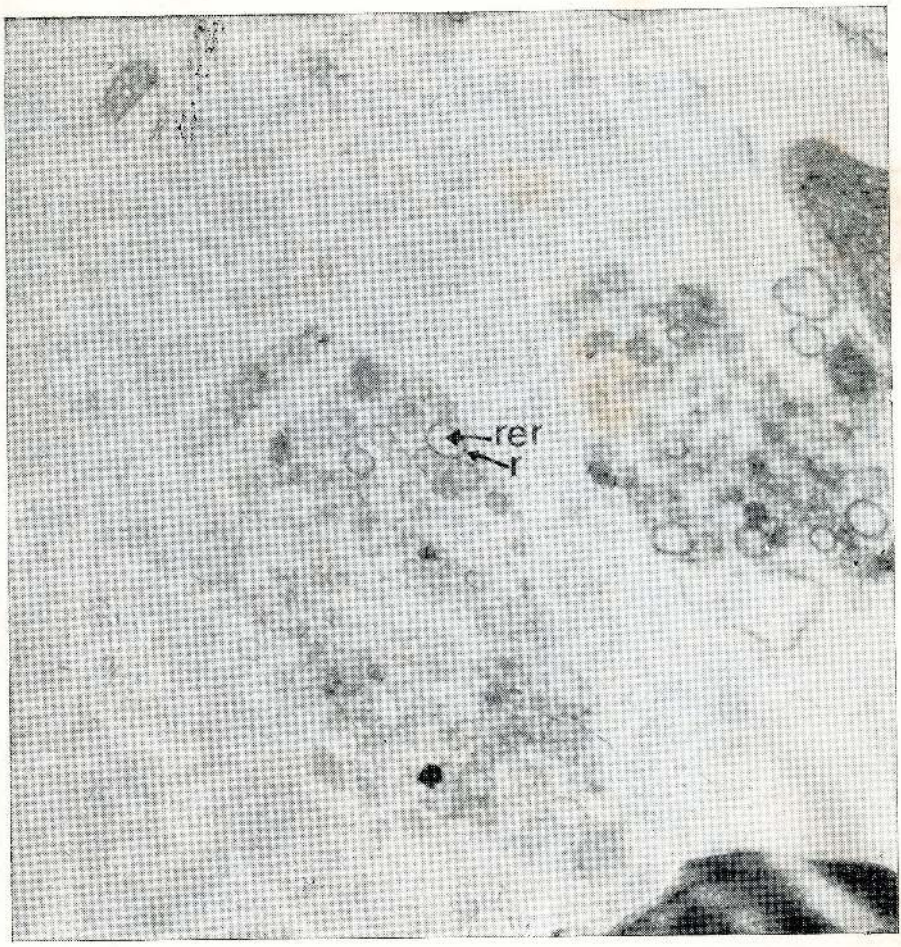


Figure 5. An electron micrograph of a hepatocyte in a tadpole moribund after four days of treatment showing distended rough endoplasmic reticulum (rer) and ribosomes (r). x 25,000

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

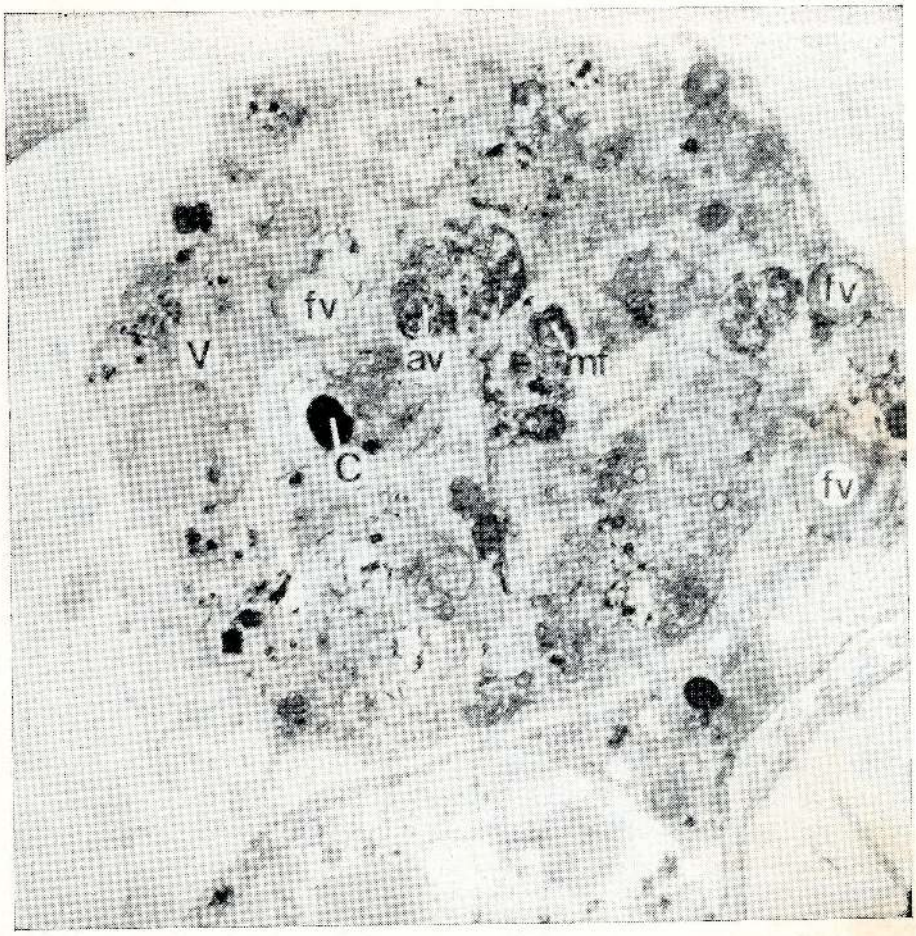


Figure 6. An electron micrograph of a hepatocyte in a tadpole moribund four days after treatment showing autophagic vacuoles (av), fatty vacuoles (fv), myelin figures (mf), vacuoles (V) and crystals (C). x 12,750

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

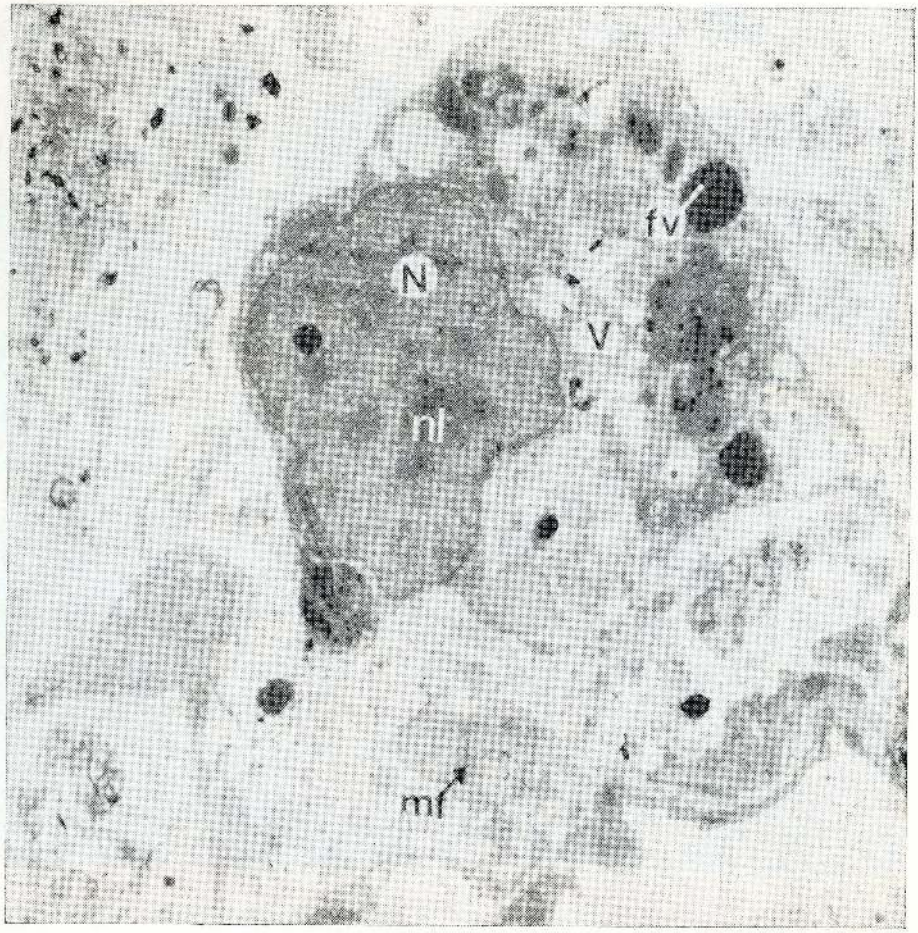


Figure 7. An electron micrograph of a hepatocyte in a tadpole moribund four days after treatment showing prominent nucleus (N), nucleolus (nl), fatty vacuoles (fv), vacuoles (V) and myelin figures (mf). x 12,800

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

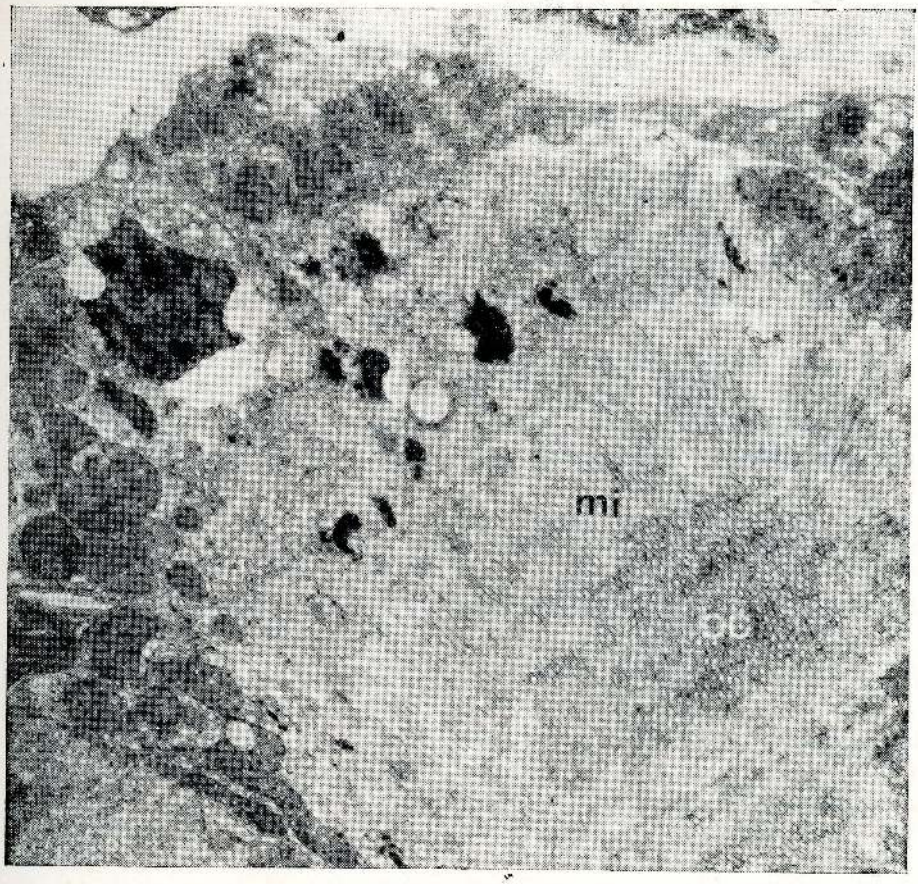
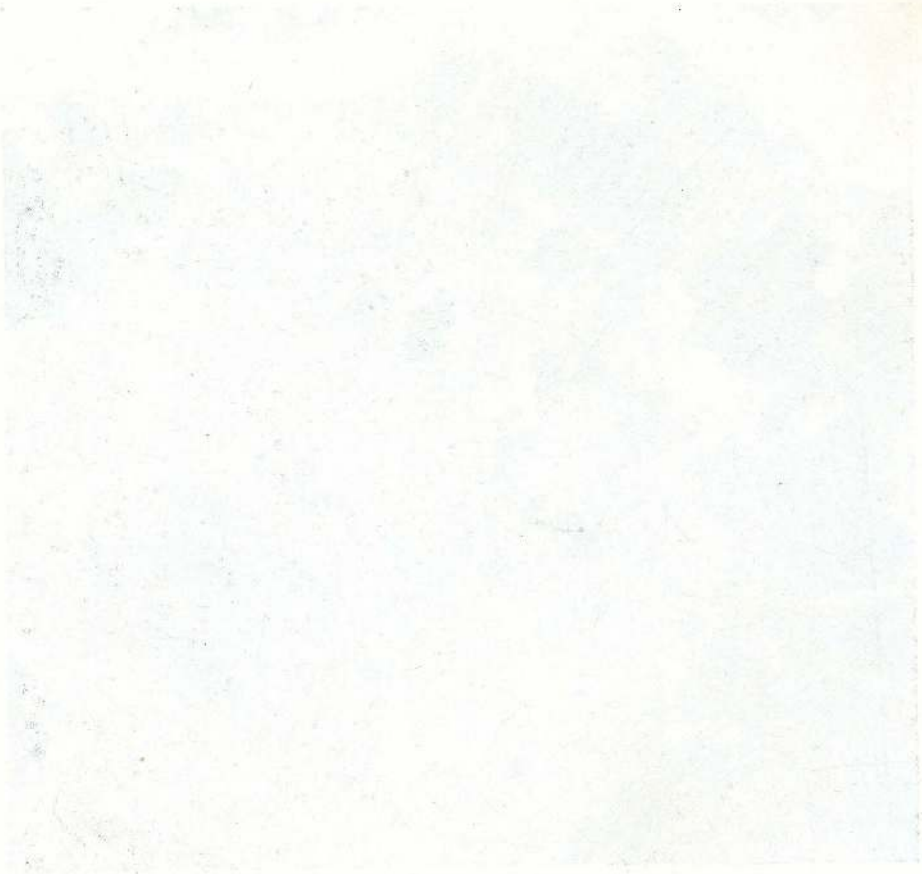


Figure 8. An electron micrograph of a hepatocyte in a tadpole moribund four days after treatment showing increased microvilli (vi), projecting into a bile canaliculus (bc). x 16,000

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media



Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

References.

1. ALLISON, A. C. & PATON, G. R. (1965). *Chromosomal damage in human diploid cells following activation of lysosomal enzymes.* *Nature* **207**: 1170 – 1173

2. ARSECULARATNE, S. N., DE SILVA, L. M., BANDUNATHA, C. H. S. R., TENNEKOON, G. E., WIJESUNDERA, S. & BALASUBRAMANIAM, K. (1969). *The use of Tadpoles of Bufo melanosticus (Schneider), Rhacophorus leucomystax maculatus (Gray) and Uperoden sp. in the bioassay of aflatoxins.* *Brit. J. exp. Path.* **50**: 285 – 294.

3. BASSIR, O. & BABAMUNMI, E. A. (1970). *Effects of anticoagulant action of aflatoxin B1 on the ultrastructure of the rat liver cell.* *Brit. J. exp. Path.* **51**: 483 – 486.

4. BUTLER, W. H. (1966). *Early hepatic parenchymal changes induced in the rat by aflatoxin B1.* *Amer. J. Path.* **49**: 113.

5. BUTLER, W. H. (1971). *Further ultrastructural observations on injury of rat hepatic parenchymal cells induced by aflatoxin B1.* *Chem. Biol. Interact.* **4**: 49 – 65.

6. JAYATILAKA, A. D. P. & MAXWELL, D. S. (1973). *An ultrastructural investigation of the effect of aflatoxin B1 on the hepatocytes of Xenopus laevis (Daudin) tadpoles.* *Proc. Cey Ass. Advmt. Sci.* **1**: 46 – 47.

7. JOSEPH, T. & JAYATILAKA, A. D. P. (1970). *Changes observed in the median eminence, the adenohyphosis and the thyroid gland in Rhacophorus leucomystax maculatus (Gray) during metamorphosis.* *Cey. J. Med. Sci.* **19**: 34 – 40.

8. SVOBODA, D., GRADY, H. J. & HIGGINSON, J. (1966). *Aflatoxin B1 injury in rat and monkey liver.* *Amer. J. Path.* **49**: 1023 – 1053.

9. SVOBODA, D. & HIGGINSON, J. (1968). *A comparison of ultrastructural changes in rat liver due to chemical carcinogens.* *Cancer Res.* **28**: 1703 – 1733.

10. THERON, J. J. (1965). *Acute liver injury in ducklings as a result of aflatoxin poisoning.* *Lab. Invest.* **14**: 1586 – 1603.

11. THERON, J. J., LIEBENBERG, N. & JOUBERT, H. J. B. (1965). *Histochemistry and electron microscopy of acute liver lesions induced by aflatoxin B1 in ducklings.* *Nature* **206**: 908 – 909.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1) :

The Utilization of Nitrogen from Spent Tea Leaf and Urea by Sheep fed Alkali Treated Rice Straw as the Sole Source of Roughage

M. C. N. JAYASURIYA

Department of Animal Science, Faculty of Agriculture, University of Peradeniya, Peradeniya, Sri Lanka.

(Date of receipt : 28 March 1980)

(Date of acceptance : 02 January 1981)

Abstract: A feeding trial with crossbred sheep, to investigate the usefulness of spent tea leaf (STL) as a source of protein for sodium hydroxide treated rice straw is reported. Three concentrate rations prepared by mixing ground maize either with urea (14 g or 18 g per 100 g concentrate) or spent tea leaf (18 g per 100 g concentrate) were compared. Treated straw (4% w/w) was given to appetite as the only source of roughage. The animals on diet containing STL performed equally well as those on urea npn diets, although STL containing diet provided only 6% crude protein. This observation points to a possible by-pass nature of STL protein. It also appears to be an excellent source of supplementary protein for sodium hydroxide treated straw-based diets.

1. Introduction.

The usefulness of non-protein nitrogen (npn) compounds in the nutrition of ruminants has been well documented. [1, 2, 12] Urea, the commonest npn compound of choice has increased the feeding value of both untreated [3], [4] and chemically treated [5, 6] poor quality roughages. Urea at 2% level of supplementation has significantly increased the digestibility and voluntary dry matter intake of sodium hydroxide treated rice straw.[8]

Spent tea leaf (STL), the residue from the manufacture of instant tea has 32% crude protein in dry matter. Feeding trials conducted in Sri Lanka have indicated the possibility of incorporating up to 18% STL in the concentrate component of rations of growing calves. [9]

The feeding trial reported in this paper was aimed at investigating the usefulness of STL as a source of protein for ruminants fed sodium hydroxide treated rice straw. An attempt was also made to compare STL protein with urea npn.

2. Materials and Methods.

The apparent digestibility of three diets was determined using growing sheep (Jaffna local x Bikaneri) of average liveweight 22 kg (range 21 kg to 24 kg). Rice straw variety H_4 chopped into 20 to 50 mm lengths, treated with 40 g sodium hydroxide (NaOH) dissolved in 1·2 litres of water per kg of straw as described previously,[7] was given to appetite as the only source of roughage. In addition, three concentrate rations (Table 1) prepared by mixing ground maize either with urea (14 g or 18 g per 100 g maize) or spent tea leaf (18 g per 100 g maize) was given at the rate of 100 g per animal per day.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 1 – Composition of concentrate rations used in the feeding trial.

	Ration Number		
	R–1	R–2	R–3
Amount of ground maize (g)	100·0	100·0	100·0
Amount of urea added to ground maize (g/100g)	14·0	18·0	–
Amount of spent tea leaf added to maize (g/100g)	–	–	18·0
Dry matter (g/100g)	89·3	88·3	90·3
Ash (g/100g dry matter)	2·9	3·3	8·5
Crude protein (g/100g dry matter)	29·8	55·6	15·5

Table 2 – The average chemical composition of treated straw, spent tea leaf and ground maize.

	treated straw	ground maize	spent tea leaf
Dry matter (g/100g)	92·4	89·8	91·8
Composition of dry matter (g/100 g dry matter) Ash	17·4	1·9	3·9
Crude protein	3·8	9·6	32·0
Crude fibre	30·8	2·3	13·4

Three animals were used for each treatment. They were housed individually in metabolism crates designed to enable the separate collection of faeces and urine. The feeding period was of 20 days duration, the first 12 days to allow for adjustment to the rations with voluntary intake and digestibility measured over the last 8 days. Voluntary intake was measured by feeding in amounts 10% greater than the previous day's intake, and determining the actual intake by daily weighing of refused feed. Water and a standard mineral mixture were freely available to all animals throughout the experiment.

Samples of concentrate, treated straw and faeces were stored for subsequent analysis for moisture, ash, crude fibre, and crude protein by conventional methods. Urine was collected daily in plastic buckets containg 16 ml of 1 N hydrochloric acid; the volume determined a 2% aliquot was refrigerated for subsequent nitrogen determination.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

3. Results and Discussion.

The average chemical composition of treated straw, ground maize and STL used in the feeding trial is shown in Table 2. Spent tea leaf used in concentrate ration R–3 had a dry matter content of 91.8% and a crude protein content of 32.0% on dry basis. The quantity of STL used in the ration was restricted to 18 g per 100 g concentrate as this level had been found to be the most suitable for growing calves in a previous study.[9]

The present observations are in general agreement with earlier reports on the influence of non-protein nitrogen supplementation on apparent digestibility of alkali treated straw.[8] Urea intake up to 2% to 3% of the total dry matter consumed increased the estimated [11] metabolizable energy value of straw to 7.5 MJ/kg dry matter, making the treated straw equivalent to a medium quality hay.

Table 3 – Intake and apparent digestibility of dietary constituents

	concentrate ration number			SE of difference between means
	R–1	R–2	R–3	
Crude protein content of total diet (g/100 g DM consumed)	9·44	12·27	6·04	–
Estimated ME content of straw[1] (MJ/kg DM)	7·75	7·74	7·61	–
Intake of urea (%) (intake as a % of total DM consumed)	2·83	2·97	–	–
Digestibility of dietary constituents (%)				
Dry matter (DMD)	62·0	61·5	61·6	0·64
Organic matter (OMD)	68·9	68·1	67·2	0·70
Digestible organic matter in dry matter (DOMD)	58·7	57·7	56·7	0·70
Derived digestibility of straw[2] organic matter	64·7	64·6	64·3	–
Intake				
Ad libitum intake of straw dry matter g/animal/day	465·5	517·0	489·1	66·13
g/Kg W 0·75/ day	48·9	54·3	51·4	6·94
N balance (g/day)	+0·99	+6·32	+1·83	–

1. Metabolizable energy estimated as DOMD X O·145 [11]
2. Concentrate organic matter digestibility assumed to be 85%

The addition of 18% STL to concentrate ration R–3 increased the crude protein content of the total diet only marginally to 6·0%, yet its influence on the apparent digestibility and voluntary intake of straw dry matter was comparable to the treatment R–2 having a crude protein equivalent of 12.3% (Table 3) Furthermore, at 6% level of crude protein this diet was able to bring about a positive nitrogen balance which in fact was twice as much as the 14% urea supplemented diet. These observations suggest a possible by - pass nature of the

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

STL protein. It is possible that the polyphenols present in STL[9] may be acting as a chemical agent in reducing microbial degradation of STL protein in the rumen, thereby making the valuable aminoacids available in the abomasum and lower digestive tract. Since the first draft of this paper was written, we have confirmed in our laboratory that the STL protein has a very low rumen solubility compared to many standard protein concentrates (example: 12% compared to 50% in the case of coconut oil meal at 8 hours of fermentation) and that the by- pass protein from STL is about 80% digestible in the lower digestive tract.

It has been established that concentrate rations prepared with up to 18% STL are highly acceptable and safe to growing calves fed forage diets.[9] A recent trial has also shown that up to 20% STL can be included in the concentrate component of dairy cattle rations without harmful effects.[8] Since supplementation of low protein feeds based on roughage with a form of by-pass protein increases feed intake and improves feed conversion ratio (kg feed intake/kg gain) of ruminants,[10] STL could become an excellent source of protein for poor quality roughage based diets, especially alkali treated straw diets.

Further research is however required to evaluate the quality of STL protein for ruminants.

Acknowledgements.

The author gratefully acknowledges the financial assistance given by the National Science Council of Sri Lanka. I am also grateful to Mr. H. G. D. Perera for technical assistance.

References

1. BALCH, C. C. (1967) *Wld. Rev. Anim. Prod.* **3** (14): 84.
2. CAMPLING, R. C. & BALCH, C. C. (1962) *Nutr. Abstr. Rev.* **32**:669.
3. CONRAD, H. R. & HIBBS, J. W. (1967) *J. Dairy Sci.*, **51**:276.
4. COOMBE, J. B. & TRIBE, D. E. (1963) *Aust. J. agric. Res.* **14**:70.
5. DONEFER, E., ADELEYE, J. O. A. & JONES, T. A. O. C. (1969) *Effect of urea supplementation on the nutritive value of NaOH treated straw.* In *Gould, R. F. (ed) Cellulases and their applications.* Am. chem. Soc., Washington, D. C.
6. JAYASURIYA, M. C. N. (1979) *J. Natn. Sci. Coun. Sri Lanka.* **7**:11.
7. JAYASURIYA, M. C. N. (1979) *Trop. Agric.* (Trinidad). **56**:75.
8. JAYASURIYA, M. C. N. (1980) *J. Natn. Sci. Coun. Sri Lanka.* **8**: 223-225
9. JAYASURIYA, M. C. N., PANDITHARATNE, S. & ROBERTS, G. (1978) *Anim. Feed Sci. Technol.*, **3**:219.
10. LENG, R. A., KEMPTON, T. J. & NOLAN, J. V. (1979) Personnal communication.
11. REID, J. J. (1953) *J. Dairy Sci.*, **36**:955.
12. UNITED KINGDOM. Ministry of Agriculture Fisheries and Food (1976) Bulletin No. 33. HMSO. London.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1) :

Salivary Immunoglobulins and Lactoferrin in Dental Caries

S. DISSANAYAKE, L. P. SAMARANAYAKE

Faculty of Medicine, University of Peradeniya, Peradeniya, Sri Lanka.

AND

R. M. BENNETT

Department of Medicine, University of Chicago, Chicago, Illinois, U. S. A

(*Date of receipt* : 07 March 1980)

(*Date of acceptance* : 15 January 1981)

Abstract: The levels of serum IgG, IgA, salivary IgA and salivary lactoferrin were determined in high and low caries subjects. The levels of immunoglobulin and lactoferrin were found to be elevated in high caries subjects.

1. Introduction.

Numerous studies have shown variations in serum and salivary immunoglobulins in dental caries, [4,7,11,15] some reports suggesting a protective function for salivary immunoglobulins. [3,4,5,9,13]

Lactoferrin, the iron and copper binding protein in secretions,[8] is present in saliva and has been described as a bacteriostatic agent.[2] As bacteria play a significant role in the establishment and progress of dental caries, it is important to know whether lactoferrin levels show any relation to the severity of disease.

In this communication, we summarise our preliminary data on serum immunoglobulins, salivary immunoglobulins and lactoferrin in dental caries in man.

2. Materials and Methods.

2·1. Subjects

The series consisted of 121 patients selected from those attending an 'Out Patients' clinic, at the General Hospital, Kandy and 28 caries free first year medical students from Peradeniya. The medical students were freshmen and were studied within one month of their arrival in the campus. Their family income was used to assess the socio-economic backgrounds and 28 students of socio-economic status comparable to that of the test group were selected. All subjects were examined by the same dental surgeon and those patients with evidence of other oral diseases, especially gingivitis, were excluded. The degree of dental caries was measured by the number of decayed, missing and filled teeth (DMF Index). The subjects were classified according to the DMF Index.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

2·2. Collection of saliva and serum.

About 5 to 10 ml of saliva were collected under paraffin stimulation from each subject. The time of saliva collection was between 10 am and 12 noon. The saliva samples were clarified by centrifugation (200 g) and stored at -20°C. Blood was collected into Haematocrit tubes by the finger prick method, serum separated and stored at -20° C.

2·3. Determination of immunoglobulin and lactoferrin levels.

Unconcentrated saliva was used in all determinations. SIgA levels in saliva were determined by Single Radial immunodiffusion (SRID) in 1.5% Agarose using a rabbit antiserum (specific) to human S IgA (Nordic Immunological Laboratories, the Netherlands). The 11S IgA standard was prepared in this laboratory. Serum IgG and IgA levels were determined by SRID in 2% Agarose using class specific antisera (Nordic Immunological Laboratories) with reference to the WHO Standard Serum 67/97.

11S IgA (as determined by gel filtration on a calibrated Sephadex G-200 column) was isolated from human colostrum. Gamma-globulins in human colostrum were isolated by repeated precipitation with 33% ammonium sulphate in phosphate buffer, pH 7.4. 11S IgA was obtained by gel filtration in the calibrated Sephadex G-200 column and further purified by affinity chromatography on CNBr-Sepharose 4B insolubilized rabbit anti- human SIgA (from Nordic immunological Laboratories, the Netherlands). The 11S IgA thus prepared showed one precipitation line on immunoelectrophoresis against rabbit anti-human Fab and rabbit anti-human SIgA (Nordic Immunological Laboratories). Calculation of standard concentrations was based on the assumed extinction coefficient for SIgA of $E^{1\,cm}_{280\,nm}\ 1\% = 13{\cdot}5$.[10] This preparation was also calibrated against the WHO Standard Serum 67/97 with a correction factor applied for differences in rates of diffusion.[14]

2·4 Determination of salivary lactoferrin levels.

Determination of salivary lactoferrin levels was performed by solid phase radioimmunoassay exactly as described previously.[1]

3. Results.

The number of subjects in each caries group, the mean $\pm$ S. D. of their age, DMF Index, levels of serum IgG, IgA, salivary IgA, and lactoferrin are shown in Table 1. In Table 2 are shown the statistical correlations (by linear regression analysis) between salivary IgA, lactoferrin and the DMF Index.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

When the DMF Index was greater than 15, the mean serum IgG levels were significantly greater than that of the control group with a DMF Index=0. The salivary IgA and lactoferrin levels showed a gradual increase with the increase of DMF Index. In high caries subjects (DMF Index = 11-15 and > 15) the mean levels of salivary IgA and lactoferrin were significantly higher than the corresponding values for the control group with DMF Index = 0.

Table 1 - The Mean, ± S. D. of Age, DMF Index Serum IgG, Serum IgA, Salivary IgA and Salivary Lactoferrin.

Subject Group	Age (years)	DMF Index	Serum IgG (IU/ml)	Serum IgA (IU/ml)	S. IgA (μg/ml/ml)	Lactoferrin (μg/ml)
DMF Index = 0	20 ± 1·2	0	57±27	51±27	67±20	5±7
(n = 28)					(46-113)	(0·2-26)
DMF Index = 1-5	21 ± 2	2·6 ± 1·2	51±21	44±26	76±36	8±11
(n = 41)					(42-176)	(0·2-41)
DMF Index 6-10	21 ± 1	7·5 ± 1·3	52±20	52±29	82±45	17±37
(n = 24)					(46-126)	(0·6 42)
DMF Index 11-15	23 ± 4	12·4 ± 1·4	54±15	64±29*	95±92*	20±21*
(n = 32)					(46-151)	(2-84)
DMF Index > 15	27 ± 6	20·9 ± 4·1	73±26**	57±22	116±113**	27±20**
(n = 24)					54-319	(5-58)

* : difference from DMF Index = 0 , significant at 20% level.
** : difference from DMF Index = 0 , significant at 0·05% level.

Table 2 - Correlation between Salivary IgA, Lactoferrin and the DMF Index

Subject Group	Salivary IgA and DMF Index	Lactoferrin and DMF Index	Lactoferrin and IgA
DMF Index = 1-5	correlation ,, —0·163 , NS	correlation ,, 0·063 NS	correlation ,, 0·062 NS
DMF Index = 6-10	,, 0·348 ,p = 0·01	,, 0·036 NS	,, 0·035 NS
DMF Index = 11-15	,, 0·281 ,p = 0·01	,, 0·4564 ,p = 0·001	,, 0·4722,p = 0·001
DMF Index = > 15	,, 0·204 ,p = 0·01	,, 0·501 ,p = 0·001	,, 0·503 ,p = 0·001

NS: not significant.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

4. Discussion.

Subjects with a DMF Index greater than 15 exhibited a significantly elevated level of serum IgG: the group with a DMF Index = 11 to 15 (but not above 15) exhibited a significant elevation of serum IgA. These findings are in general agreement with other reports.[4, 7] In contrast, salivary IgA and lactoferrin demonstrated a gradual increase with the increase of the DMF Index. The standard deviations were so high, however, that only in the group with DMF Index greater than 15 (greater than 10 for lactoferrin) was the difference from the control group sufficient to be statistically significant. To our knowledge, this is the first report on elevated levels of lactoferrin in dental caries.

The concentration of salivary proteins are known to be affected by the rate of saliva secretion.[3, 10] However, it is not known to what extent these experimentally determined differences reflect true differences in the glandular secretion of salivary proteins. In the present investigation, we were not able to measure the rates of saliva secretion and therefore, it was not possible to correct for differences in rates of flow.

The absolute levels of salivary IgA observed in the present series are higher than the values reported by others.[4,6,10,12,15] Such variations are to be expected for reasons of ethnic, social and economic differences. However, technical errors due to differences in Standard SIgA preparations cannot of course be excluded. Unfortunately an international standard for SIgA is not available and therefore accurate standardizations could not be made.

The pattern of variation of salivary IgA in relation to the DMF Index probably reflects the stimulation of salivary IgA production by antigens of cariogenic organisms. A deficiency of secretory immunoglobulins in saliva precipitating dental caries seems unlikely. The significance of the observed elevation of salivary lactoferrin in high caries subjects is not clear. Further studies to elucidate the role of lactoferrin in dental caries and also the antibody specificity of salivary IgA are in progress.

References

1. BENNETT, R., M. & MOHLA, C (1976) *J. Lab. clin. Med.* **88** : 156.
2. BULLEN, J. J., ROGERS, H. J. & LEIGH, C. (1972) *Br. Med. J.* **1** : 69.
3. BRANDTZAEG P. (1976) *J. dent. Res.* (Special issue) **55** : c 102.
4. CHALLACOMBE S. J. (1976) *Caries Res.* **10** : 165.
5. COLE, M. F., ARNOLD, R. R., RHODES, M. J.& McGHEE, J. R. (1977) *J. dent. Res.* **56** : 198.
6. EVERHART, D. L., GRIGSBY, W. R. & CARTER, W. H. (1972) *J. dent. Res.* **51** : 1487.
7. LEHNER, T., CARDWELL, J. E. & CLARRY, E. D. (1967) *Lancet* **1** : 1294.
8. MASSON, P. C. & HEREMANS, J. F. (1966) *Protides Biol. Fluids Proc. Collog Bruges.* **14** : 115.
9. MICHALEK, S. M., McGHEE, J. R., MESTECKY, J., ARNOLD, R. R. & BOZZO, L. (1976) *Science* **192** : 1238.
10. ORSTRAVIK, D.& BRANDTZAEG, P. (1975) *Archs. oral Biol.* **20** : 701.
11. SHKLAIR, I. L., ROVELSTED, G. M. & LAMBERTS, B. C. (1969) *J. dent. Res.* 48 : 842.
12. SHILLITOE, E. J. & LEHNER, T. (1912) *Archs. oral Biol.* **17** : 241.
13. TAUBMAN, M. A. & SMITH, D. J. (1974) *Infection Immunity* **9** : 1079.
14. TOMASI, J. B. & GREY, H. M. (1972) *In "Structure & function of immunoglobulin A". Prog. Allergy.* **16** : 18-213. *Karger, Basel.*
15. ZENGO, A. N., MANDEL, I. D., GOLDMAN, R. & KHURANA, N. S. (1971) *Archs. oral Biol.* **16** :557.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1):

Impetigenised Scabies and Acute Glomerulonephritis in Sri Lanka; a Prospective and Retrospective Study

S. N. ARSECULERATNE, N. CHARAVANAPAVAN, C. NAVARATNAM

Department of Microbiology, Faculty of Medicine, University of Peradeniya, Peradeniya, Sri Lanka.

AND

D. A. GUNAWARDENE

Dermatology Unit, General Hospital, Kandy, Sri Lanka.

(Date of receipt : 24 March 1980)

(Date of acceptance : 23 January 1981)

Abstract : From a study of patients, hospitalized for acute glomerulonephritis with impetigenised skin lesions, impetigenised scabies was found to be the commonest antecedent infection (79%); this confirms reports from Trinidad and South Africa that impetigenised scabies is a common precedent of acute glomerulonephritis. A prospective study of patients with impetigenised scabies showed that only 8% developed urinary abnormalities. Possible explanations for this difference are discussed. The pattern of the serotypes of the β haemolytic streptococci isolated from these skin lesions was similar to that reported from other countries. *Staphylococcus aureus* alone was recovered from the skin lesions of 40% of patients with acute glomerulonephritis (AGN) but the wide variation of the properties of these isolates did not suggest the existence of special nephritogenic strains. Amongst the isolates of *S. aureus* from the skin lesions of patients with or without AGN, phage Group III was the commonest, with phage Type 54 predominating. The staphylococcal isolates differed in their phage pattern distribution, from those reported from temperate countries. AGN following pyoderma, was found to have a latent period similar to that reported from other countries; the extent of the pyodema had no relation to the occurrence or severity of the AGN. The ASO titres in the nephritics, following pyoderma were found to be intermediate between those in normal persons and those in acute rheumatic fever. Epidemics of post-streptococcal AGN have not been experienced in this country although sporadic cases are common.

1. Introduction

Acute glomerulonephritis (AGN) as a sequel to a β haemolytic streptococcal infection, has been well documented.[10] In temperate climates, especially in the winter and in spring, the streptococcal infection has most often been an upper respiratory tract infection. In warmer seasons, in these countries and in tropical climates, pyoderma is reported to have been the more common antecedent.[10] This is particularly true of populations living under poor hygienic conditions.

The pathogenesis of the renal lesion of AGN has generally been recognised as being due to immune complexes and the acute nephritic syndrome which typically includes post-streptococcal AGN has also been described in association with other systemic diseases including staphylococcal[23, 28], viral[2] and parasitic infections.[24]

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

AGN and acute rheumatic fever (ARF) are two major clinical problems in this country and in 1969/1970 the yearly admissions to state hospitals for ARF and AGN were approximately 450 and 760 per 100,000 population, respectively. In 1976, the rates for ARF and AGN were 500 and 720 respectively.

It has been a clinical impression that many of our AGN patients, especially in the paediatric age group, have associated scabies complicated by pyogenic superinfection, which is one of the commonest dermatological conditions seen in this country. In 1969/1970 the admission rate to state hospitals for the treatment of scabies was 4500, while in 1976, the rate was 1990 per 100,000 population. These figures exclude an appreciable number of cases that would have sought outdoor or traditional indigenous treatment.

Since there was no data on the association between AGN and scabies in this country, we studied the inter-relationships between scabies, a supervening β haemolytic streptococcal or staphylococcal infection of the skin, and AGN, both prospectively by following up patients who presented with scabies and retrospectively in patients with AGN, in this district over a one and a half year period-1969/1970- to take into account, seasonal variations in rainfall and temperature. The topics of this study included the Groups and Types of the streptococci, the characteristics of the strains of *Staphylococcus aureus* isolated from the skin lesions and the anti-streptolysin titres (ASOT) in normal persons and in the patients.

2. Experimental

The study populations: All patients were of a low socio-economic status, attending the non-fee levying state hospital in this district. They were consecutive cases divided into the following classes:

Class A had 85 patients (48 male and 37 female) with AGN and impetigenised skin lesions (infected scabies, infected eczema, infected dermatomycosis and impetigo), who were warded in the medical and paediatric wards of this hospital. AGN was diagnosed when any three of the following were present-albuminuria, oedema, hypertension, macroscopic haematuria or microscopic (more than 5 red cells per high power field in the deposit from centrifuged urine) and oliguria.

Class B had 18 patients with AGN without preceding or concomitant pyoderma.

Class C had 55 patients (38 males and 17 females) who formed the prospective study group. They presented with infected scabies at the Dermatological unit of this hospital. They were examined weekly for at least 6 weeks for symptoms and signs of AGN. Scabies which was in all cases secondarily infected was diagnosed clinically[32] by a contact history, pruritus, distribution of the lesions

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

and the response to antiscabetic therapy. Microscopic examination for the parasite was not done at this stage on account of the difficulty of identifying the burrows in the pigmented skin and the isolation of the parasite with secondary infection. The clinical examination, as in the patients of Classes A and B, included a detailed examination of the cardiovascular system, the urinary tract with a laboratory examination of the urine as described below. The clinical examination included a measurement of the blood pressure after a short period of rest especially in outdoor patients. A standard blood pressure chart was used to assess the significance of the measured pressure. A fresh sample of urine was examined for albumin by the acid–heat method and for deposits microscopically. The cell count was expressed as their number per high power field (X 400) in the deposit from 5 ml of urine centrifuged at 150 g for 10 min. The throat and at least 10 skin lesions were swabbed with serum coated swabs which were plated immediately. A sample of peripheral venous blood was taken for ASOT determination.

After the examination, the patients were treated appropriately, with 25% benzylbenzoate emulsion, for scabies, with or without oral or parenteral penicillin or tetracycline.

Normal subjects. Sera from 176 normal school children (age range 5–15, geometric mean 6.4; 82% males, 18% females) were used as controls for the ASOT study.

Bacteriological methods. For primary isolation, the throat and skin swabs were plated on 5% sheep blood agar with and without crystal violet and the plates were incubated aerobically and anaerobically. β haemolytic streptococci, after purification were grouped by testing acid extracts by gel diffusion against standard grouping sera ('Wellcome Diagnostic Reagents'). Typing of Group A strains was done by Dr Jiri Rotta (Institute of Epidemiology and Microbiology, Prague, Czechoslovakia).

The strains of *S. aureus* were characterised as follows:- haemolysis on sheep blood agar, pigmentation on milk agar, lipolysis on ('Oxoid') tributryin agar, opacity production on egg yolk agar, DNAse production on ('Difco') DNse test agar, mercuric chloride sensitivity[22] penicillinase production[15], antibiotic resistance using standardised discs ('Rosco' Sensitabs, Denmark) of penicillin, tetracycline and streptomycin with the 'Oxford' staphylococcus as reference strain; independent confirmation of the zone diameters recommended by the manufacturers was made by comparison with the frequency distribution of zone diameters (Arseculeratne, unpublished data) of 300 strains of *S. aureus* isolated in this laboratory and by MIC determinations with pencillin in tube dilution tests. The strains were phage typed with the following phages :-

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Group I - 29, 52, 52A, 79, 80

Group II - 3A, 3B, 3C, 55, 71

Group III- 6, 7, 42E, 47, 53, 54, 75, 77, 83A

Group IV - 42D

Miscellaneous - 8I, I87

Strains untypable by these phages were also tested against the following additional phages :- 84, 85, 88.

Antistreptolysin 'O' titres:- these were estimated with commercial kits ('Wellcome Diagnostic Reagents') by the 50% endpoint method determined colorimetrically, as recommended by the manufacturers.

3. Results

Class A. The age distribution of the patients with AGN in Class A was as follows, indicating that AGN in this country is primarily a paediatric problem:-

age (years)	number of cases	approximate %
1–5	16	19
6–10	36	42
11–15	11	13
16–20	11	13
over 20	12	14

Nature of the skin lesion. The nature of the impetigenised skin lesions in the 85 patients with AGN was as follows:-

lesion	number of cases	approximate %
infected scabies	67	79
impetigo	14	16
infected eczema	3	4
wound infection	1	1

Although the lesions in all the scabetic patients appeared to be infected with pyogenic organisms, bacterial isolation was successful in only 63 (74%) of cases. 20 patients had been given systemic antibacterial therapy before bacterial culture was attempted. In relation to prior penicillin or tetracycline treatment, the isolation of β haemolytic streptococci from the skin lesions was successful in 5/24 treated and 24/61 untreated patients.

Area of skin affected. Although assessment of the area of the affected skin was imprecise, there appeared to be no correlation between the area of the skin involvement and the occurrence of AGN or its severity.

Latent period. In 79 cases, the modal latent period between the onset of the scabies and the AGN was 4 weeks with a mean of 9·3 weeks.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 1 – The incidence of β haemolytic streptococcus and *Staphylococcus aureus* in the impetigenised skin lesions of patients in Class A and Class C.

Class	β haemolytic streptococcus		S. *aureus*		Both β haemolytic streptococcus and S. *aureus*	neither
	skin	throat	Skin	throat	skin	
A	12	1	34	4[a]	17	22
C	13	2[a]	17	4[a]	18	11

b same strain as from skin lesions
a strains different from those from skin lesions
Figures refer to number of cases

Table 2 – The Phage pattern and Group distribution of *Staphylococcus aureus* from the impetigenised skin lesions of patients in Class A (numbers of strains within parenthesis)

phage Group				
I	II	III	Mixed	untypable
29 (4)	55 (2)	54 (8)	29/54 (1)	24
29/52/52A/79/80 (1)	55/71 (2)	53/54 (2)	71/42E (1)	
29/71 (1)	3A/55/71 (1)	77 (1)		
	71 (1)	6/53/54/83A (1)		
	79/3C/55/71/81 (1)			
Total 6	7	12	2	24

Table 3 – The properties of *Staphylococcus aureus* isolated from the skin lesions of patients in Class A.

Phage Group						colour[a]			P'nase[b]		Lipo[c]		EY[d]		Hg[e]		AB resistance[f]			
I	II	III	IV	Misc	NT*	GY	C	W	+	—	+	—	+	—	S	R	I	2	3	nil
6	9	12	0	0	24	24	25	2	10	41	43	8	43	8	49	2	10	5	0	36

*untypable, a—GY golden yellow, C cream, W white, b—penicillinase, c— lipolysis, d— Egg yolk
e— mercury resistance, f— antibiotic resistance to 1, 2, 3 or nil antibiotics

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Identity of the organisms. The incidence of β haemolytic streptococci and *S. aureus* is shown in Table 1. The group and type distribution of the streptococci are shown in Table 5 and 6 respectively. None of the patients had more than one strain.

The properties of the staphylococci are shown in Table 3 and are similar to those of Class C. All the strains were positive for coagulase (slide and tube) and DNase production. As in Class C, the commonest phage Group was III. The phage patterns of 51 strains are shown in Table 2.

The phage group distribution of *S. aureus* in Class A and C was similar to that in a separate series of 110 strains of *S. aureus* which were isolated from skin lesions in this hospital, in which the distribution was as follows :-

Group	number of strains
I	16
II	10
III	23
IV	1
Misc	2
nontypable	58

The commonest phage types in order of frequency were 54 (Group III), 79 (Group II) and 55/71 (Group II). *Class B.* 18 patients with AGN had no history of impetigenised scabies or other skin lesions and no detectable skin lesions on examination. 8 patients (44%) gave a history of sore throat with an average latent period of 3.3 weeks. β- haemolytic streptococci type T 22 and T 44 were isolated from the throat in only two patients (12%) respectively. Throat cultures were negative for *S. aureus*. *Class C.* The incidence of β haemolytic streptococci and *S. aureus* in the skin lesions and in the throat of the 55 patients is shown in Table 1.

Only 50 patients turned up for follow-up examination for the 6 week observation period. The five patients who were not followed up, kept in touch by post and reported cure of their skin lesions and absence of clinical evidence of AGN. Of the 50 patients who were followed up weekly, four (8%) showed microscopic haematuria, with albuminuria but none of these patients developed the clinical picture of overt AGN (Table 4).

The group and type distribution of the strains of β haemolytic streptococci isolated from the skin lesions in patients of Class C are shown in Tables 5 and 6 respectively.

The properties of the strains of *S. aureus* isolated from the skin lesions of these patients are shown in Table 7. Of the 55 patients, 35 had *S. aureus* in their skin lesions, either alone or with β haemolytic streptococci. None had more than one type of *S. aureus*. All these strains were coagulase positive

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 4 – The characteristics of patients of Class C who developed urinary abnormalities after impetigenised scabies.

age (years)	duration of scabies (weeks)	area affected %	organism in skin	week of albuminuria or haematuria	ASOT
2	2	12	*Staph. aureus*[1]	1	ND*
11	4	5	β haemolytic streptococcus T 9	1	ND*
14	?	3	nil	1	200
18	4th recurrent attack	8	nil	1,2	710

1 cream coloured, phage untypable, mercury sensitive, EY –, Penicillin resistant
* not determined

Table 5 – The Group distribution of strains of β haemolytic streptococcus isolated from the skin and throat in patients with pyoderma and AGN (Class A), the throat in patients with AGN without pyoderma (Class B) and from patients with impetigenised scabies (Class C)

Lancefield Group	Class A skin	Class A throat	Class B throat	Class C skin	Class C throat
A	23	1	2	26	2
C	1	0	0	1	0
G	2	0	0	4	0
Total	26	1	2	31	2

Table 6 – The Type distribution of Group A strains of β haemolytic streptococci isolated from the skin and throat of patients with pyoderma and AGN (Class A), the throat of patients with AGN without pyoderma (Class B) and from patients with impetigenised scabies (Class C)

Broad range Type classification of T Type	Class A skin	Class A throat	Class B throat	Class C skin	Class C throat
5/11/12/27/44	3	0	0	6	0
3/13/B3264	3	0	0	8	1
8/25/Imp 19	6	0	0	2	0
14/35/49	1	0	1	4	0
15/17/19/23/40/47	0	0	0	1	0
4/24/26/28/29/46	1	0	0	1	0
M55 T25/Imp 19	2	0	0	1[a]	0
T 22	4	1	1	0	1[b]
T 9	1	0	0	2	0
untypable	2	0	0	1	0
Total	23	1	2	26	2

a. the only strain typable by the M antigen
b. strain different from that isolated from the skin

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

by the slide and tube tests and produced DNase. There was a total positive correlation between the EY and lipolysis reactions. The phage patterns in each group were as follows :–

Group I	Group II (number of strains	Group III within parentheses)	Miscellaneous
79 (2)	55/71 (4)	54 (6)	187 (1)
29 (2)		53 (2)	
80 (1)		(83A 1)	
29/79 (1)			
79/80 (1)			
Total 7	4	9	1

A minor outbreak of AGN in a small hamlet.

The hamlet consisted of a closed community of three families which lived in wattle and daub houses, adjacent to one another. The children were siblings or cousins and were in constant and intimate contact. The outbreak occurred during the rainy month of July. Four children were admitted to hospital within two days for AGN and infected scabies. They had all received parenteral penicillin from their rural hospital before admission. The remaining 5 children had no symptoms of AGN but all had impetigenised scabies. These five were treated by us for their skin infection and were followed up weekly for 3 months. None developed the urinary abnormalities or clinical evidence of AGN.

In the children with AGN, the ages ranged from 8 months to 10 years and they had scabies for 1 to $1\frac{1}{2}$ months. No streptococci were isolated from their skin lesions

In the contact children whose ages ranged from 7 months to 7 years, the duration of scabies was from 1 to 2 months with a mean of 1·5 months. The isolates of β haemolytic streptococci from these five contact children had the following serotypes :-

case	site	T antigen	Broad range Type classification
2	throat	5111/12/27/44	5/11/12/27/44
	skin	12	5/11/12/22/44
2	skin	11/12	5/11/12/27/44
	skin	12	5/11/12/27/44
3	skin	5/11/12/27/44	5/11/12/27/44
4	skin	4	4/24/26/28/29/46
5	skin	non - typable	—

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

The strains of *S. aureus* isolated from the skin lesions in all the children, nephritic and contact, were identical in all the properties tested-golden yellow coloured, phage type 54 (Group III), positive reactions in the following tests - DNase production, tube and slide coagulase production, egg yolk opacity production, lipolysis, sensitivity to mercuric chloride and penicillin.

Table 7 – The properties of *Staphylococcus aureus* isolated from the impetigenised scabetic lesions of patients in Class C

Phage Group				colour[a]			P'nase[b]		Lipo[c]		EY[d]		Hg[e]	antibiotic[f] resistance			
I	II	III	Misc.	NT*	GY	C	W	+	–	+	–	+	–	S	R	1	2	3	nil
7	4	9	1	15	15	17	3	11	23	28	7	28	7	32	3	12	4	1	18

*untypable, a – CY golden yellow, b – penicillinase, c – lipolysis, d – Egg yolk
C cream
W white
e – mercury resistance, f – antibiotic resistance to 1, 2, 3 or nil antibiotics

Table 8 - ASOT in normal subjects and in patients with AGN with pyoderma (Class A), patients with AGN without pyoderma (Class B), patients with impetigenised scabies (Class C)

Subjects	number	ASOT range	ASOT mean	mode
normal	176	50 — 1600	354[a] ± 342 233[b] (inappropriate)	170
Class A	59	123 — 1600	760[a] ± 420 668[b] (appropriate)	640
Class B	9	141 — 1600	799	
Class C	14	200 — 1448	644	

[a] arithmetic mean, [b] Geometric mean, Davies' test was used to assess the appropriateness of either mean to the values in the two classes.

Antistreptolysin 'O' titres.

Data concerning the ASOT in normal subjects and in patients with AGN (with and without pyoderma) is shown in Table 8. The cumulative frequency distribution of ASOT in normal subjects is shown in Figure 1.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

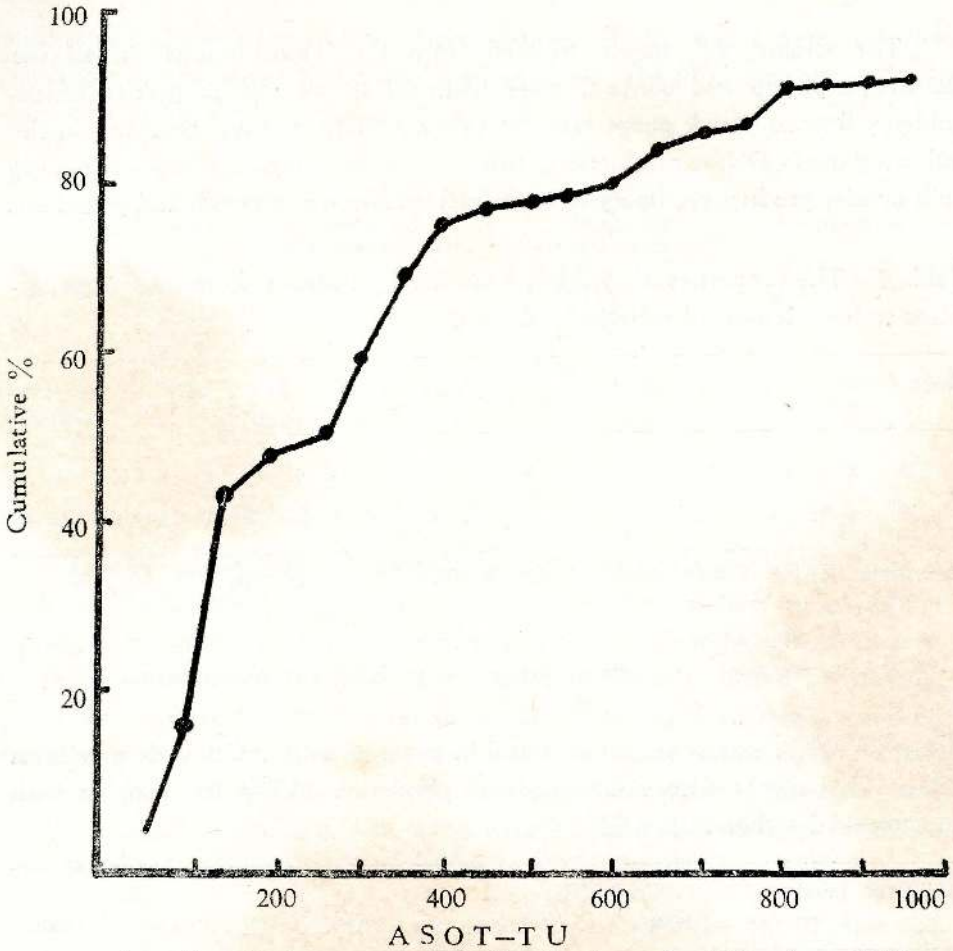


Figure 1. Cumulative percentage distribution of ASO titres (TU) in 176 normal persons

The mean titres in the normal subjects were significantly different from those of Class A ($p < 0.001$), of Class C ($p < 0.05$) and those of 9 patients with ARF ($p < 0.001$); the patients with ARF were hospitalised during this period and their ASOT ranged from 640 to 1600 with a mean of 1112. The mean ARF titre differed significantly ($p < 0.001$) from those of Class A and Class C.

There was no significant difference between the titres of patients Class C and those in either Class A or B. There was also no significant difference between the titres in patients with and without recoverable β haemolytic streptococci from their skin lesions in Class A and in Class C.

4· Discussion

In temperate countries, the streptococcal infection which precedes AGN has commonly been an upper respiratory tract infection.[30] In warmer climates however, the commonest antecedent infection has been streptococcal pyoderma[10]

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

and recently, pyogenic infection of scabies was documented as a common precedent of AGN in South Africa[14] and in Trinidad.[32] It was stated[5] that over half the children in the Red Lake epidemic of AGN in the Indian Reservation of Minnesota were suspected of having had scabies. Allen[1] described AGN which followed a β haemolytic streptococcal infection on scabetic lesions in a patient in a temperate country in winter. Our finding of the preponderance of impetigenised scabies as the antecedent in AGN in Sri Lanka confirms these observations. In addition, the broad parallelism between the incidence of scabies and AGN in this district (Figure 2) is compatible with this relationship. Hence the early treatment of scabies, which Hersch[14] recommended be classified as a notifiable disease, may be a useful step in reducing the incidence of AGN.

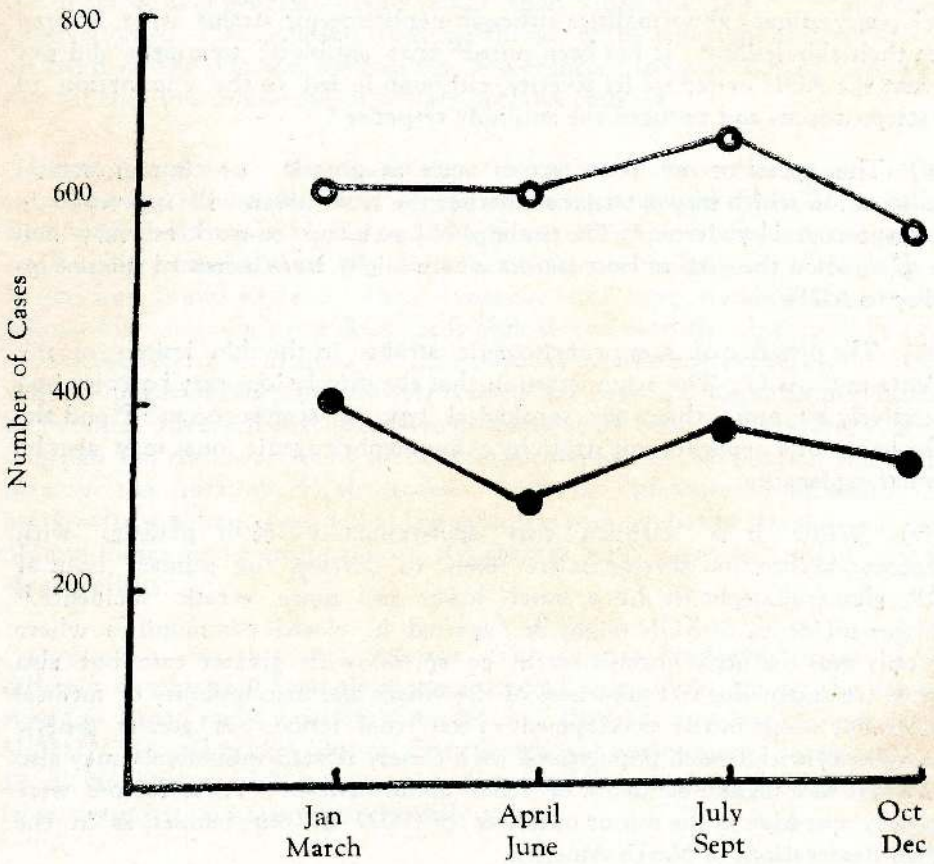


Figure 2. Quarterly admissions for the treatment of Acute Glomerulonephritis (-●-) and of Scabies (-o-) in the General Hospital, Kandy, Sri Lanka: the numbers represent means of quarterly admissions for the years 1969 - 1972.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

The incidence rate of streptococcal pyoderma in the nephritics of Class A is in contrast to the lower rate of development of urinary abnormalities suggestive of AGN in patients who had similar skin lesions and a pattern of streptococcal and staphylococcal colonisation, in the prospective series of Class C. Other prospective studies too have recorded relatively low rates for the development of urinary abnormalities. [5, 6, 18, 21] Hersch[14] found evidence of AGN in 8% of 100 outdoor patients who had scabies. Svartman and co-workers[32] found urinary abnormalities, haematuria and proteinuria, in 9% of scabetics on follow up. Several factors could account for this difference between prospective and retrospective studies :-

(i) the treatment of the pyoderma before the patients were seen by us, and the suppression of the streptococcal colonisation and hence the antistreptococcal immune response. This was perhaps the case in those patients who did not develop any urinary abnormalities although nephritogenic strains were isolated from their skin lesions. It has been noted[20] that antibiotic treatment did not prevent the AGN or reduce its severity, although it led to the elimination of the streptococcus and reduced the antibody response.[4]

(ii) The operation of host factors such as genetic or immunological predisposition which may determine whether the renal lesion will supervene on the streptococcal pyoderma. The findings of Lasch and co-workers[20] may also be explained on the basis of host factors which might have increased the susceptibility to AGN.

(iii) The presence of non-nephritogenic strains in the skin lesions of the patients in Class C. The demonstration that the skin lesions may be colonised successively by more than one serological type of streptococcus[5, 26] and the replacement of a nephritogenic strain by a non-nephritogenic one may also be another explanation.

(iv) While it is claimed that approximately 3% of patients with streptococcal infection develop or are likely to develop the primary bout of ARF, glomerulonephritis has a much lower and more erratic incidence.[29] A higher incidence of AGN might be expected in closed communities where not only may the nephritogenic strain be spread with greater ease but also delayed treatment due to remoteness of the village and inaccessibility of medical centres may result in the development of the renal lesion. A greater genetic homogeneity within such populations with closely related individuals may also contribute to a higher incidence of renal abnormalities. These factors were probably operative in the minor outbreak of AGN in our hamlet, as in the Indian Reservations of North America.

Latent period. The average latent period of clinically overt AGN following streptococcal pyoderma is generally reported as being between 1-4 weeks.[10] The modal latent period in our nephritics was 4 weeks. It is significant that microscopic haematuria was observed within the first 2 weeks of pyoderma in

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

4 patients of Class C. Raffel[29] commented "patients who will subsequently develop nephritis are likely to show some red cells in the urine during the acute stage of the streptococcal infection". It is however difficult to say whether the microscopic haematuria would have progressed to overt AGN in the absence of therapy or follow up. Renal tubular lesions have been described as a reaction to the 'S' streptolysin of the β haemolytic streptococcus[33] including non-nephritogenic strains, and to *S. aureus*.[28] Hence in the absence of renal biopsy in cases not showing clear clinical signs and symptoms of AGN, it is difficult to attribute the haematuria to AGN.

Source of streptococci. It has been concluded from studies in other countries[7, 10, 11] that respiratory colonisation with the same strain of streptococcus as on the skin occurs subsequently to the skin infection. Isolation rates from the pharynx have generally been lower than from the skin.[21, 32] It was our observation too that isolation from the throat, of the same type of streptococcus as found on the skin was less frequent and occurred in only 2 out of 55 prospective patients and in one out of 85 retrospective patients

Extent of pyoderma. We found no correlation between the extent of the pyoderma and the occurrence or length of the latent period of AGN although Hersch[14] found that the incidence of AGN was roughly related to the extent and severity of the skin lesions. In some of our nephritics, only a few scabetic lesions were found while in others extensive and long standing lesions were followed by clinically overt AGN only after several months. Infection by non-nephritogenic strains in some of these episodes may be one explanation. Other patients may have been inadequately treated for scabies. Kaumheimer quoted by Futcher[12] observed that there was no correlation between the severity of the impetigo and the character of the subsequent nephritis. No relation was found between the duration of the scabies and the presence of albuminuria.[14] Markowitz *et al*[21] also found that the duration and severity of the skin infection did not differ in the nephritics in comparison with patients without urinary abnormalities.

Streptococcal serotypes in pyoderma. The relative isolation rates of the different Groups of β haemolytic streptococcus were similar among the Classes A and C. Broad range types 8/25/Imp 19 and Type 22 in Class A and Types 5/11/12/27/44 and 3/13/B 3264 in Class C were the commonest. Types 8/25/Imp 19, 3/13/B 3264 and 5/27/44 were also described[9, 10] as common serotypes isolated from streptococcal impetigo with AGN.

Two out of 22 strains from the skin lesions of nephritics in Class A and one out of 33 strains from patients with pyoderma (Class C) were of M Type 55. Rotta (1970, personal communication) referred to this type as having also been recognised as a new type in other tropical countries. M Type 55/T8/25/Imp 19 has also been isolated from cases of AGN in Trinidad[26, 27] and in Israel.[20]

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Most of our pyoderma strains were untypable by the M antigen, an experience previously recorded.[9, 18, 21, 36] The pattern of streptococcal serotypes isolated from our cases as pyoderma conformed to the impetigo serotypes described by other workers [2, 5, 6, 16, 18, 25], especially in our finding that common isolates were of the serotypes 8/25/Imp 19,, 3/13/B 3264 and 5/27/44. The Red Lake serotype M49 was however absent amongst our isolates from cases of AGN.

Staphylococcus aureus. The pattern of staphylococcal isolates from the skin lesions however differed from those reported by other workers.[6, 21, 25] Phage type 71 (Group II) which they found to be the commonest type in impetigo, occurred in only 5/51 of our Class A and 4/21 Class C strains. In both Class A and Class C of our series gave phage Group III and I as the predominant groups with phage type 54 (Group III) as the commonest type; phage type 54 was also the commonest type which we recovered in a separate study of 110 strains (unpublished data) from skin infections. Group III strains were also found to be the commonest in pyoderma in American Indian children.[5]

S. aureus was isolated more frequently than β haemolytic streptococci from the skin lesions in both prospective and retrospective patients in our series, an experience also reported by other workers.[6, 21] The latter authors suggested from ASO titres in skin infections, that streptococci may be more frequently associated with pyogenic skin infections than isolation rates would suggest and that staphylococcal infection is perhaps secondary. The finding that *S. aureus* phage type 71, a common impetigo type, inhibits the growth of β haemolytic streptococci[8] may explain the failure to recover the streptococcus from mixed infections.

The pathology of the renal lesions following staphylococcal septicaemia has been described in detail;[28] lesions similar to those of post-streptococcal AGN such as exudation and glomerular proliferation, were seen. Diffuse glomerular lesions indistinguishable from those of post-streptococcal AGN have been described.[23] The characteristics of the staphylococcal strains in Powell's study[28] were not described. In our series, the strains of *S. aureus* isolated from the nephritics of Class A did not fall into any well defined group in terms of biochemical properties, phage types or antibiotic sensitivity patterns. This finding is in contrast to the occurrence of special nephritogenic types of the β haemolytic streptococcus. Our finding, with regard to the isolation and serotypes of the β haemolytic streptococcus which are similar to those reported by other workers, supports the pathogenic role of certain strains in the development of AGN. However the absence of any clear pattern of properties, at least among those tested, of the isolates of *S. aureus* from the nephritics, would suggest the absence of particular 'nephritogenic' strains of this species.

The fact that the strains of *S. aureus* isolated from the minor outbreak were identical not only in phage type but also in all the other properties tested, reflects the ease of spread of a single strain within this community in which the individuals were in close contact.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Epidemic AGN. Widespread epidemics have been reported from the USA[10], in the Red Lake Indian reservation and from Trinidad.[32] "Whether the nephritis follows skin sepsis or infection of the respiratory tract, it tends to appear in epidemics because only some strains of Group A streptococcus are nephritogenic".[3] It is possible that the strain type 5/11/12/27/44 also spread to all the patients in our hamlet, as did the strain of *S. aureus* but the isolation of the streptococcus from the skin lesions of the nephritic children was probably prevented by the penicillin treatment which these patients received before they were examined by us. This type was present in the nephritics of Class A as well and was also isolated by Dillon[10] from skin lesions in patients with AGN. As stated above, the occurrence of outbreaks of AGN in closed communities such as our hamlet and in the Indian reservations of North America may be contributed to by host factors such as genetic homogeneity as well as by close contact of the individuals

Apart from this outbreak, we have not encountered epidemics of AGN in this country. However we do have seasonal increases in the incidence of sporadic cases. The infrequency of epidemics is perhaps also reflected in the multiplicity of serotypes of the streptococci in our cases.

Antistreptolysin titres. Gunatillake and Perera,[13] from a study of normal children under 12 years living in a semi-urban area in Sri Lanka reported that the greatest number of children showed titres between 100 and 166 TU. Our finding of a mode of approximately 150 TU in this age group agrees with their figure. They found that 29·5% of their children under 12 years had titres of over 166 although in our series for the same age group, 56% had titres over 150 TU. A possible explanation for this higher value could be that our study population was of a lower socio-economic status with more overcrowding in their living conditions and a higher incidence or severity of streptococcal infections. Variations of ASOT tend to occur with age, geographic location, the frequency of streptococcal infections, within a given location and with population groups.

The upper level for the normal population in the US (80% of subjects having titres at this level or lower) was 333 for the 5–12 year group and 200 for the young adults.[34] Koshi and Mammen[19] reported an upper level of 333 for normal children between 6–15 years in South India, while Markowitz and co-workers[21] considered a level of 250 for this age group in the USA.

The ASO titres in our nephritics with pyoderma showed lower values than did the patients with ARF, as has been reported from other countries.

Acknowledgements

The authors gratefully acknowledge the assistance in this study of Dr. Jiri Rotta (International Reference Centre for Streptococcus typing, Prague, Czechoslovakia) who serotyped the streptococci, of Dr. Yvette Hermon and Mr. K. Kuganesan, (Virus Laboratory, Colombo South Hospital, Sri Lanka) who phage typed the staphylococci and of the staff of the Paediatric wards of the General Hospital, Kandy, Sri Lanka.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

References

1 ALLEN, B. R. (1972) *Lancet*, **1**: 434

2 ANONYMOUS (1968) *Brit. med. J.*, **3**: 264

3 ANONYMOUS (1968) *Lancet*, **1**: 575

4 ANONYMOUS (1974) *Lancet*, **2**: 390

5 ANTHONY, B. F., PERLMAN, L. V. & WANNAMAKER, L. W. (1967) *Pediatrics*, **39**: 263

6 BARROW, G. I. (1955) *J. Hyg.Camb.*, **53**: 495

7 BLUMBERG, R. W. & FELDMAN, D. B. (1962) *J. Pediat.*, **60**: 677

8 DAJANI, A. S. & WANNAMAKER, L. W. (1971) *J. exp. Med.*, **134**: 588

9 DILLON, H. C. Jr., MOODY, M. D. & MAXTED, W. R. (1966) *J.Pediat.*, **69**: 982

10 DILLON, H. C. Jr. (1967) *Ann. Rev. Med.*, **18**: 207

11 FERRIERI, P., DAJANI, A. S., CHAPMAN, S. S., JENSEN, J. B. & WANNAMAKER, L. W. (1970) *New Engl. J. Med.*, **283**: 832

12 FUTCHER, P. H. (1940) *Arch. int. Med.*, **65**: 1192

13 GUNATILLAKE, P. D. P. & PERERA, T. D. S. (1970) *J. Hyg. Camb.*, **65**: 13

14 HERSCH, C. (1967) *S. Afr. med. J.*, **41**: 29

15 HOLT, R. J. & STEWART, G. J. (1963) *T. clin. Path.*, **16**; 263

16 JOHNSON, JENNIFER C. & STOLLERMAN, G. H. (1969) *Ann. Rev. Med.*, **20**: 315

17 KOSHI, GRACE, MAMMEN, K. C., FELDMAN, D. B., BHAKTHAVIZIAM, C. & MYERS, RUTH M. (1967) *Ind. J. med. Res.*, **55**: 920

18 KOSHI, GRACE, WEBB, J. K. G. & MYERS, RUTH M. (1968) *Ind. J. med. Res.*, **56**: 951

19 KOSHI, GRACE, & MAMMEN, K. C. (1969) *Ind. J. med. Res,*, **57**: 1347

20 LASCH, ELI E., FRANKEL, V., VARDY P A. BERGNER RABINOWITZ, S., OFEK, I. & RABINOWITZ, K. (1971) *Lancet*, **1**: 475

21 MARKOWITZ, M., BRUTON, H. D., KUTTNER, A. G. & CLUFF, L. E. (1965) *Pediatrics*, **35**: 393

22 MOORE, B. (1960) *Lancet*, **2**: 453

23 NYDAHL, B. L. & HALL, W. A. (1965) *Ann. int. Med.*, **63**: 27

24 OSBORNE, C. A., HAMMER, R. F., STEVENS, J. B., RESNICK, J. S. & MICHAEL, A. J. (1977) *Adv. in vet. Sci. comp. Med.*, **21**: 207

25 PARKER, M. T., TOMLINSON, A. J. H. & WILLIAMS, R. E. O. (1955) *J. Hyg. Camb.*, **53**: 458

26 PARKER, M. T., BASSETT, D. C. J., MAXTED, W. R. & ARNEAUD, J. D. (1968) *J. Hyg. Camb.*, **66**: 657

27 POTTER, ELIZABETH V., MORAN. A. F., POON KING T. & EARLE, D. P. (1968) *J. lab. clin. Med.*, **71**: 126

28 POWELL, D. B. (1961) *J. Path. Bact.*, **82**: 141

29 RAFFEL,S. (1961) *Immunity*, 566, 567 p. 2nd Ed. New York: Appleton, Century & Crofts.

30 RAMMELKAMP, C. H. Jr., WEAVER, R. S. & DINGLE, J. R. (1952) *Trans. Assoc. Amer. Phys.*, **21**: 598

31 ROTTA, JIRI (1970) Personal communication

32 SVARTMAN, M., POTTER, E. V., FINKLEA, J. F., POON KING, T. & EARLE, D. P. (1972) *Lancet*, **1**: 249

33 UNANUE, E. R. & DIXON, F. J. (1967) *Adv. in Immunol.*, **6**: 1

34 WANNAMAKER, L. W. & AYOUB, E. M. (1960) *Circulation.*, **21**: 598

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1) :

Preliminary Laboratory Studies on Eppawela Apatite

J. AMARASEKERA, R. SOORIYAKUMARAN AND M. G. M. U. ISMAIL

Ceylon Institute of Scientific and Industrial Research (CISIR), P. O. Box 787, Colombo 7, Sri Lanka

(Date of receipt : 18 August 1980)

(Date of acceptance : 23 January 1981)

1. Introduction

Eppawela apatite deposit was discovered during systematic geological mapping of the Anuradhapura area in North Central Province of Sri Lanka by the Geological Survey Department in 1971. This deposit spread around an area of 3 sq miles, but was most predominantly concentrated in an area of 576 acres in the northern region of the deposit and most of the investigations have been carried out in this region

This is a firm reserve of 25 million tons of apatite having P_2O_5 content of 30% to 35%. The inferred reserve for the entire deposit is around 40 million tons and it is estimated to be about 30 million tons of 90% apatite material, and it will last for 500 years at present demand rate.

Eppawela rock phosphate by itself can only be used as a fertilizer for long term crops. It cannot be directly applied to short term crops since it releases phosphate very slowly. For short term crops superphosphate is used. It is projected that in 1980 the country will require 50,000 tons of concentrated superphosphate and 46,000 tons of rock phosphates.[3]

Chemical analysis of Eppawela apatite deposit was carried out by both local and foreign institutions, and it is given in Table 1.

Table 1 – Chemical Analysis of Eppawela Apatite (From ref. 4)

		percentage by weight
CaO	—	55·30
P_2O_5	—	40·75
SrO	—	1·18
SiO_2	—	0·14
Fe_2O_3	—	0·07
MnO	—	0·01
MgO	—	0·01
F	—	1·78
Cl	—	2·29
		101·53
Less O, F, Cl	—	1·27
		100·26

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Though the P_2O_5 content of Eppawela apatite is higher than that of imported superphosphate to Sri Lanka (Table 2), the citric solubility is low when compared to other types of phosphates (Table 3).

Table 2 – Comparison of major constituents between Eppawela Apatite and imported superphosphate

Constituent	Eppawela Apatite % (by wt)	Imported Super-Phosphate % (by wt)
SiO_2	0·1 — 0·5	5·0 — 6·0
P_2O_5	30 — 35·0	28·0 — 29·0
Fe_2O_3	1 — 4	1 — 2
CaO	50 — 55	47
Cl	3·0	3·0
F	1·7 — 2·4	2·8

Table 3 – Comparison of citric acid soluble P_2O_5 in Eppawela Apatite and other fertilizers

Fertilizer Material	Citric Acid Solubility % P_2O_5
Eppawela Apatite	1·0 — 3·0
Imported rock phosphate	3·4
Imported super phosphate	9·0
Rhenania phosphate	18·9

Thus, although the citric acid solubility which is used as an index of the effectiveness of rock phosphate to the soil of the local material is about 50% less than imported phosphate, it is not suitable to use as a direct fertilizer. Further a report from Tennessee Valley authority[6] states that the Eppawela apatite is not recommended for direct application and is not considered suitable for superphosphate manufacture unless beneficiated. Hence research work had been carried out in Sri Lanka on beneficiation of Eppawela apatite.[1, 5, 8]

Since Eppawela apatite contains an appreciable amount of Cl (as well as F), it can cause corrosion in wet process beneficiation. Besides, sulphuric acid has to be imported for use in the manufacture of superphosphate fertilizer. Therefore the beneficiation method, that is most suitable for Sri Lanka would be a dry process.

To achieve this goal the Mineral Technology Section of the Ceylon Institute of Scientific and Industrial Research had carried out a series of laboratory experiments on Eppawela Apatite to produce an effective fertilizer by using locally available materials as far as possible, in a dry process.

Calcination experiments were carried out at high temperatures with local minerals such as quartz, dolomite, feldspar, normal salt and with alkali salts like soda ash, hydrated lime, sodium hydroxide and also with paddy hull ash. It was found that beneficiation with soda ash gave a product having about

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

27·00% to 30·00% of P_2O_5 in citric acid soluble form. Hence this product which is actually a thermophosphate can be used for short term crops of Sri Lanka instead of imported phosphate fertilizers by saving valuable foreign exchange which our country needs for other development work. Field trials are presently being carried out to evaluate the effectiveness of this product as a fertilizer.

The main differences between this product and Rhenania type[12] fertilizer which is made out of apatite, soda ash and silica are the method of preparation and temperature of calcination. This product is made by calcining apatite with soda ash alone at a low temperature of 1150°C than that of Rhenania phosphate (1300°C to 1400°C). Also, this has higher citric acid solubility than Rhenania phosphate.

2. Experimental

In all the experiments, the powdered samples brought from the factory at Eppawela were used. Treatments were carried out in platinum crucibles with duplicates in temperature controlled electric furnaces. Treated samples were quenched in air rapidly by immersing the crucible in a water surface. In some fusions that were carried out to find the quenching effect, experiments with sodium carbonate, quenching was done in water by immersing rapidly the platinum crucible with hot sample in water. Calcination studies of samples were carried out by using weight ratios of apatite and material at a given condition as given in respective tables. Each sample was analysed for its 2% citric acid solubility according to official methods of analysis A O A C.[2] P_2O_5 content was determined by a spectrophotometric method using ammonium vandate reagent.[9] DTA analysis of raw apatite was done by using "Spektromom 190 A" derivatograph and X-ray analysis of samples were done by using "JEOL JDX-8S" X-ray powder diffractometer.

Initially a preliminary analysis was carried out on apatite rock by DTA, X-ray diffractometer and chemically. Later fusions were carried out at high temperatures (above 1000°C) with locally available minerals and alkali salts.

Cell dimensions of sodium carbonate fused apatite samples were determined from X-ray powder diffraction patterns, taken using Cu target under following conditions, scanning ¼°/min, sollar slit 1° divergence slit1° receiving slit 0·4 mm, time constant 2sec. For this purpose peaks due to (211) and (300) planes of samples were taken.

3· Results and Discussion

3·1 Preliminary Investigation

3·1·1 *Chemical Analysis*

Chemical analysis showed the powdered rock sample which was used for these experiments has 34·00% of total P_2O_5 out of which only 9·70% is in citric acid soluble form (citric acid soluble P_2O_5 3·30%).

3·1·2 *X-ray Analysis*

X-ray analysis showed Eppawela apatite is mainly in the fluorapatite form with small amounts of chlorapatite, hydroxyapatite, α quartz, goethite (Fig 1 (A).

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

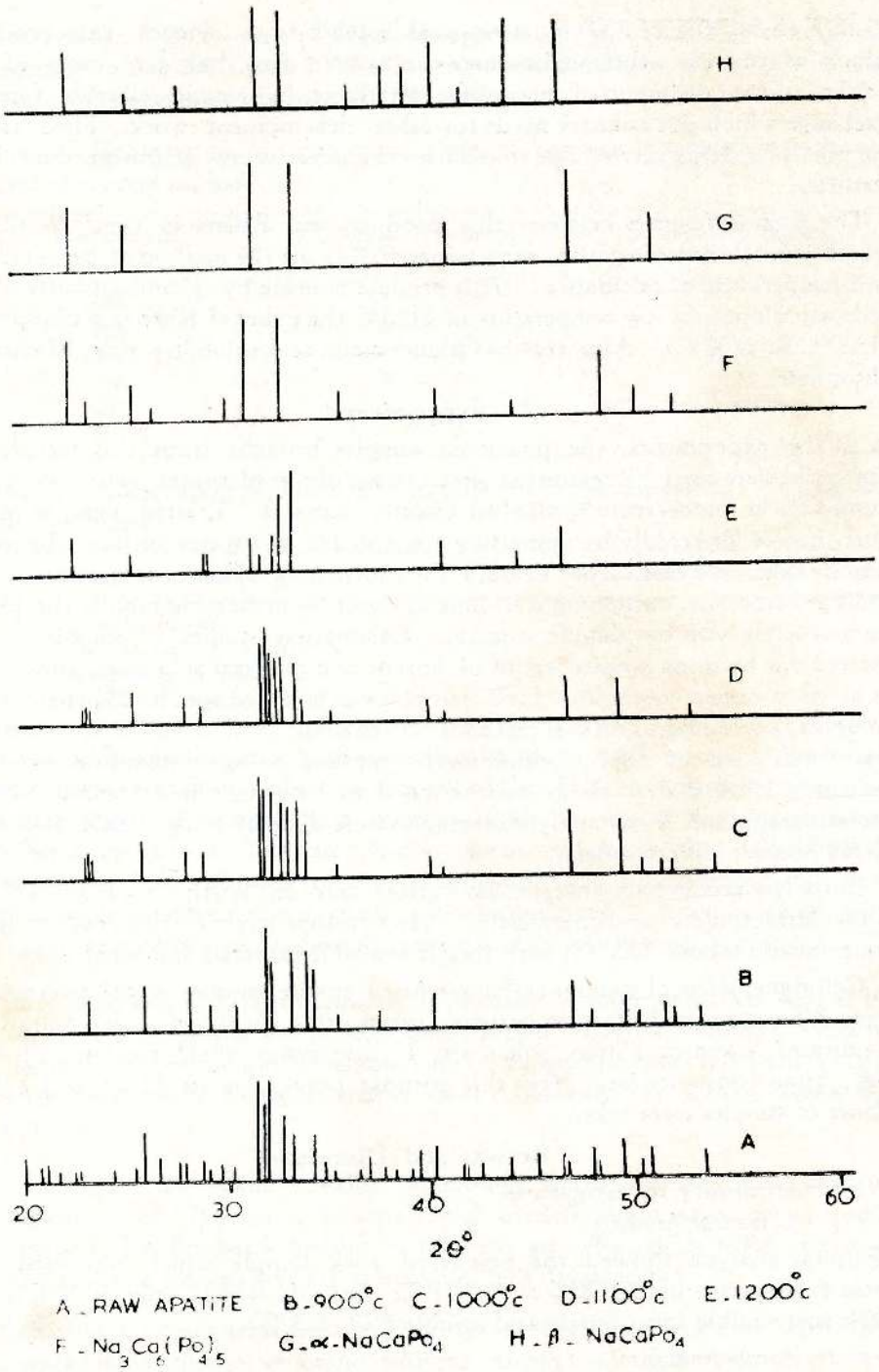


Figure 1 XRD patterns of Row Apatite and Calcined Apatite samples with Na_2CO_3 in the ratio 100 : 20 for 3·0 hrs duration.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Apart from the peaks of the above mentioned constituents, several other shifted apatite peaks were observed. These may be due to frankolite ($Ca_5(PO_4\ CO_3\ OH)_3\ F$) a polymorph of fluor-chlor and hydroxy-apatite. A firm conclusion cannot be achieved due to the unavailability of X-ray data of fran-

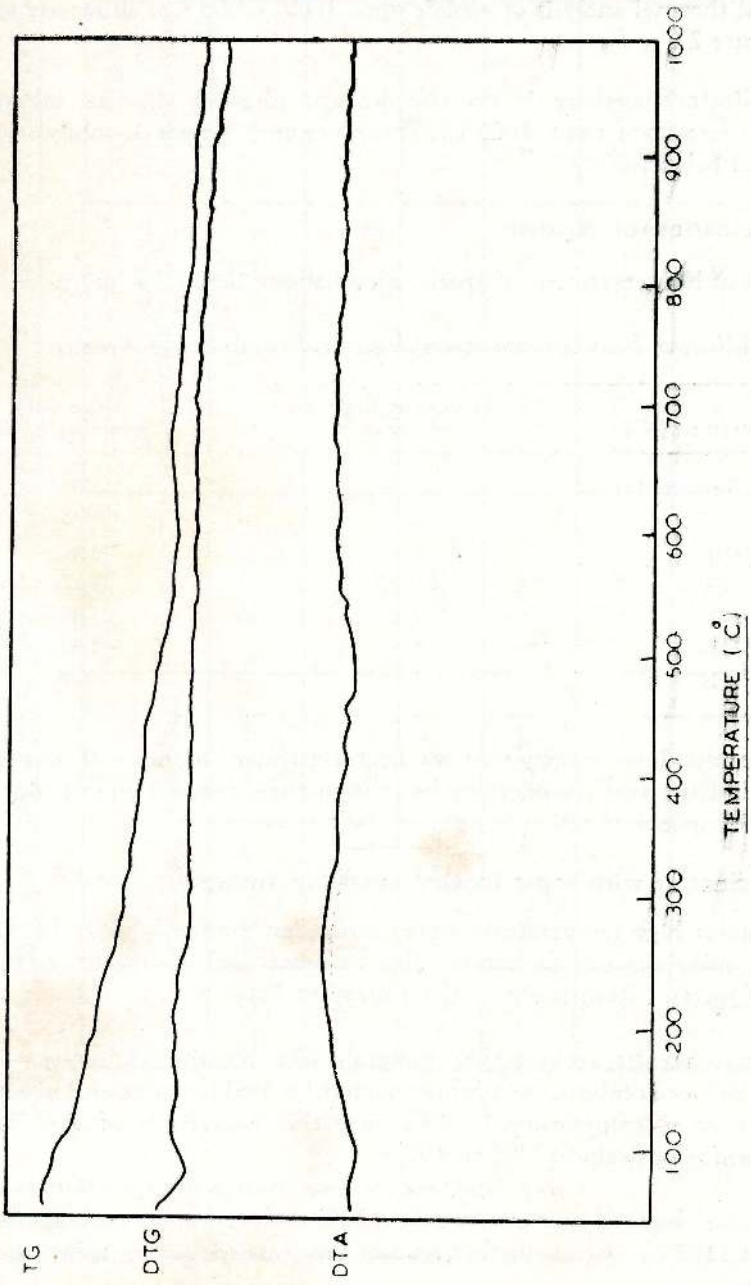


Figure 2 – DTA of Apatite

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

kolite. By taking X-ray peaks d = 2·828 A° (211) and d = 2·704 A° (300), lattice constants of Eppawela apatite was calculated and found to be a=9·4572 A° and b = 6·6786 A°.

3·1·3 *Differential Thermal Analysis*

Differential thermal analysis of apatite upto 1000° C did not show any apparent peaks (Figure 2).

This indicates that there is no chemical or physical changes taking place during heat treatment upto 1000°C. Therefore any changes in solubility cannot be expected below 1000°C.

3·2 Calcination of Apatite

The results of heat treatment of apatite alone above 1000°C is given in Table 4

Table 4 – Effect of heat treatment on citric acid solubility of Apatite

Temperature (°C)	Treatment duration (hrs)	citric acid solubility% P_2O_5
Room Temperature	—	3·33
1000	0·50	3·35
1100	0·50	3·40
1200	0·50	3·60
1000	1·0	3·40
1100	1·0	3·52
1200	1·0	3·75

These results shows clearly that the heat treatment alone will not increase solubility in citric acid considerably by changing the temperature or duration of treatment of apatite.

3·3 Calcination with some locally available minerals

Calcinations at high temperatures were carried out randomly with the following powdered minerals; (a) Dolomite (b) Feldspar (c) Kaolinite (d) Normal Salt (e) Quartz. Results obtained are given in Table 5.

The above results reveals that minerals like Kaolin, Dolomite, Feldspar, Common Salt or combination of these cannot be used for successful beneficiation of apatite even at temperatures 1100°C, since the conversion of P_2O_5 for citric soluble form is only about 17% to 19%.

It was also observed that quartz gave a 30% conversion on rock apatite when calcined at 1100°C. Hence further studies were carried out by using quartz as a mineralizer.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

TABLE 5-EFFECT OF CITRIC ACID SOLUBILITY OF APATITE WHEN CALCINED WITH DIFFERENT MINERALS

RATIO			TEMPERATURE OF TREATMENT °C	DURATION OF TREATMENT Hrs	CITRIC ACID SOLUBLE P_2O_5 %	% CONVERSION P_2O_5 TO CITRIC SOLUBLE FORM
APATITE	MINERAL(1)	MINERAL(2)				
100	DOLOMITE (30)	KAOLINE (20)	1000	2,00	6,00	17,64
100	DOLOMITE (40)	QUARTZ (20)	1200	2,00	6,00	17,64
100	FELDSPAR (40)	"	1100	2,00	6,50	19,11
100	SALT (30)	QUARTZ (20)	1100	2,00	6,50	19,11
100	QUARTZ (10)	-	1100	2,00	10,40	30,58
100	QUARTZ (20)	"	1100	2,00	10,00	29,41

3·4 Calcination with Quartz and Paddy hull Ash

The results obtained by calcination of apatite samples with quartz at different temperatures and for different time duration are given in Table 6, 7 and 8.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 6 – Effect of temperature of treatment on citric acid solubility of Apatite when calcined with constant amount of quartz for constant time duration

RATIO		Temp. °C	Duration (hrs)	Citric acid solubility % P_2O_5	% Conversion of P_2O_5 to citric acid soluble form
Apatite	Quartz				
100	10	1000	2·00	8·00	23·52
100	10	1100	2·00	10·40	30·58
100	10	1150	2·00	11·40	33·52
100	10	1200	2·00	11·50	33·82

Table 7 – Effect of duration of treatment on citric acid solubility of Apatite when calcined with constant amount of quartz at constant temperature.

RATIO		Temp. °C	Duration (hrs)	Citric acid solubility % P_2O_5	% Conversion of P_2O_5 to citric acid soluble form
Apatite	Quartz				
100	10	1150	2·00	11·40	33·52
100	10	1150	2·50	12·00	35·29
100	10	1150	3·00	12·10	35·58

Table 8 – Effect of amount of quartz used on citric acid solubility of Apatite when calcined at constant temperature for constant time duration.

RATIO		Temp. °C	Duration (hrs)	Citric acid solubility % P_2O_5	% Conversion of P_2O_5 to citric acid soluble form
Apatite	Quartz				
100	0	1150	2·50	3·80	11·17
100	10	1150	2·50	12·00	35·29
100	20	1150	2·50	11·40	33·52
100	30	1150	2·50	9·50	27·94

The above results show that by increasing temperature of treatment, about 35% of conversion can be obtained. Increasing of quartz percentage in treatments shows a lowering of citric acid soluble P_2O_5 percentage. Also it shows that about 2 to 3 hrs. treatment (Table 7) is sufficient for complete reaction to take place, between quartz and apatite.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Further experiments were carried out by using paddy hull ash obtained by burning paddy hull at 650°C. This paddy hull ash contains about 95% silica in the amorphous state. The results obtained are given in Table 9, 10 and 11.

Table 9 – Effect of temperature of treatment on citric acid solubility of Apatite when calcined with constant amount of paddy hull ash for constant time duration

RATIO		Temp. °C	Duration (hrs)	citric acid solubility % P_2O_5	% Conversion of P_2O_5 to citric acid soluble form
Apatite	PHA				
100	30	1000	2·00	6·50	19·11
100	30	1100	2·00	8·00	23·52
100	30	1200	2·00	10·00	29·41
100	10	1000	2·00	10·50	30·88
100	10	1100	2·00	11·00	32·35
100	10	1150	2·00	11·50	33·82
100	10	1200	2·00	11·60	34·11

Table 10 – Effect of duration of treatment on citric acid solubility of Apatite when calcined with constant amount of Paddy hull Ash at 1150°C

RATIO		Temp. °C	Duration (hrs)	Citric acid solubility % P_2O_5	% Conversion of P_2O_5 to citric acid soluble form
Apatite	PHA				
100	10	1150	2·0	10·50	30·88
100	10	1150	2·5	12·00	35·29
100	10	1150	3·0	12·20	35·88

Table 11 – Effect of Apatite : Paddy hull Ash ratio used for calcination on citric acid solubility

RATIO		Temp. °C	Duration (hrs)	Citric acid solubility % P_2O_5	% Conversion of P_2O_5 to citric acid soluble form
Apatite	PHA				
100	10	1150	2·50	12·00	35·29
100	20	1150	2·50	11·40	33·52
100	30	1150	2·50	10·50	30·94
100	10	1100	2·0	11·00	32·35
100	30	1100	2·0	8·00	23·52
100	50	1100	2·0	8·00	23·52
100	60	1100	2·0	9·00	26·47
100	100	1100	2·0	8·00	23·52

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

From the above results obtained on calcination experiments of apatite with quartz and PHA, effect of temperature of treatment, effect of duration of treatment and effect of ratio of constituents used for treatments are plotted (in Figures 3, 4 and 5) and variation of citric acid solubility is clearly seen.

From these results it is clear that:

a) By increasing the temperature and duration of treatment or by varying the amount of apatite to material used the highest possible citric soluble P_2O_5% that can be obtained is 12·0%, irrespective of silica material used (whether quartz or paddy hull ash). That is percentage conversion of P_2O_5 in rock apatite is 35·29%.

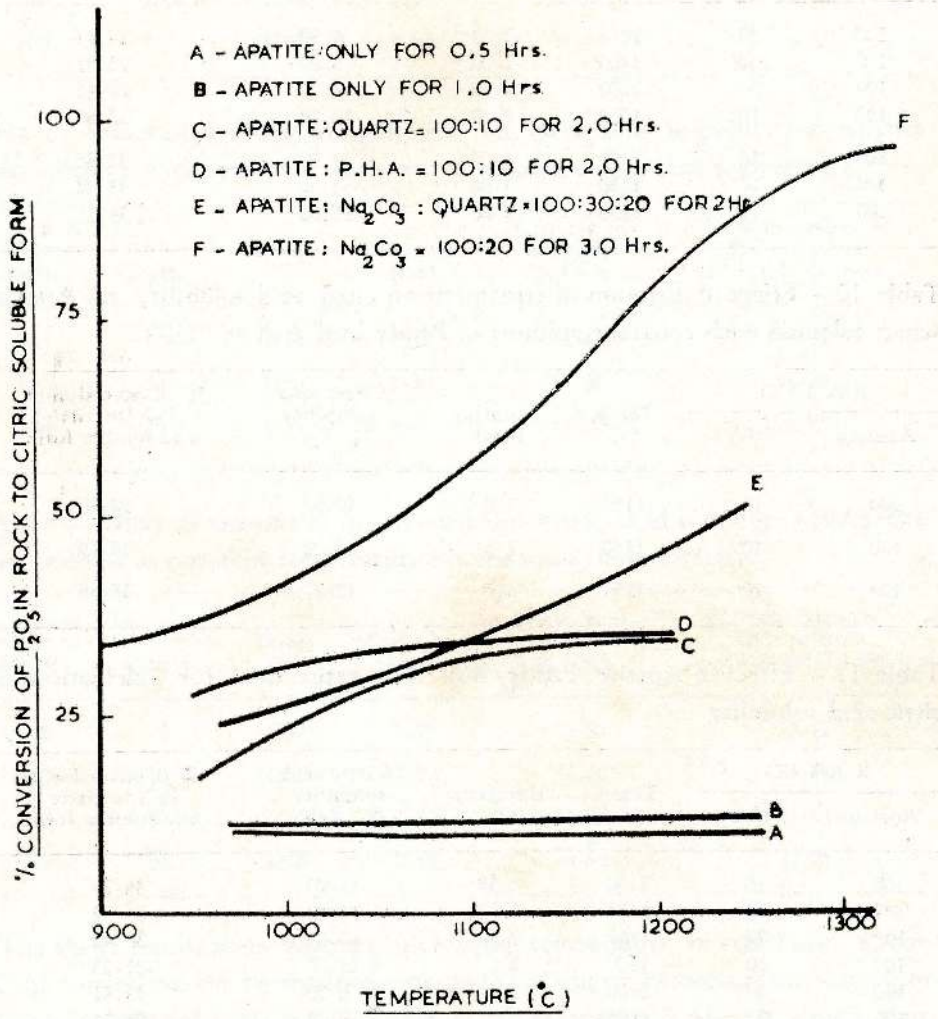


Figure 3 — Effect of temperature on percentage conversion of P_2O_5 in Apatite to Citric Acid soluble form when calcined with different mineralizers.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

b) To attain complete reaction between Paddy hull Ash or Quartz with apatite at high temperatures 2·50 to 3·00 hrs is sufficient (Figure 4).

c) From Table 8 (and Figure 5) it is clear that there is an optimum apatite : Paddy hull Ash or quartz ratio that gives the highest citric soluble P_2O_5%.

d) According to Figure 3 it is clear that we could not expect a higher citric soluble P_2O_5 % than the maximum value obtained (12·00%), by increasing temperature of treatment above 1200°C.

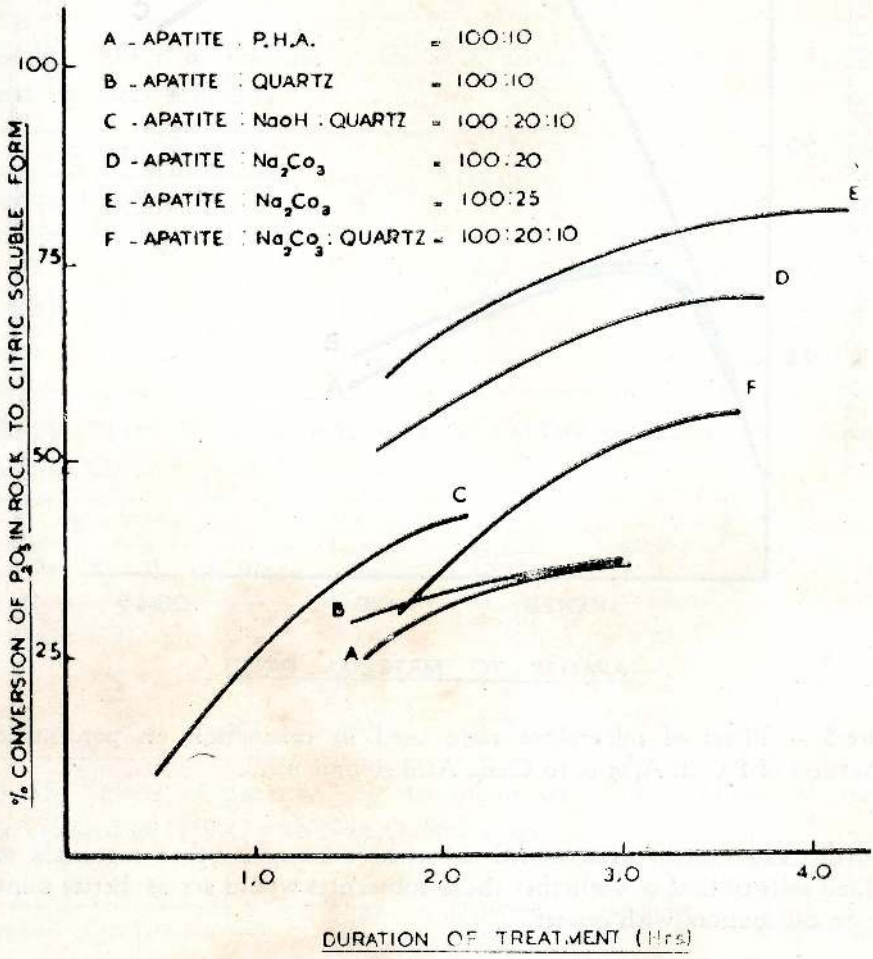


Figure 4 — Effect of duration of calcination on percentage conversion of P_2O_5 in Apatite to Citric Acid soluble form when calcined with different mineralizers.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

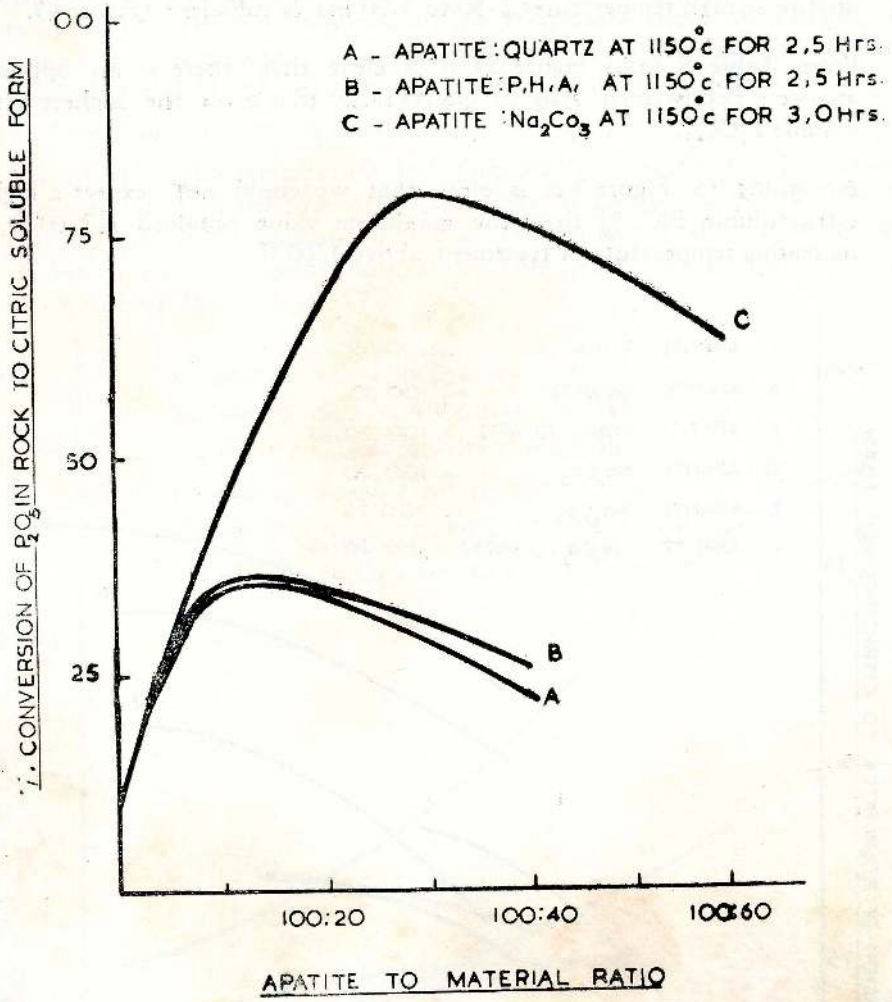


Figure 5 — Effect of mineralizer ratio used in calcination on percentage conversion of P_2O_5 in Apatite to Citric Acid soluble form.

Further experiments were carried out with quartz and other materials such as alkali salts to find out whether those substances would act as better mineralizer on calcination with quartz.

3.5 Calcination with alkali compounds

Calcinations were carried out with apatite and quartz along with NaOH, $Ca(OH)_2$ and Na_2CO_3. Following are the results obtained: Table 12, 13, 14 and 15.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 12 – Effect of NaOH on apatite : quartz calcination Effect of Duration of treatment on citric acid soluble P_2O_5%

RATIO			Temp. °C	Duration (hrs)	Citric acid solubility $\%P_2O_5$	% Conversion of P_2O_5 to citric acid soluble form
Apatite	Quartz	NaOH				
100	10	20	1000	0·50	3·40	10·00
100	10	20	1000	1·00	10·40	30·58
100	10	20	1000	2·00	13·40	39·41

Table 13 – Effect of amount of $Ca(OH)_2$ used in calcination of Apatite : Quartz on citric solubility

RATIO			Temp. °C	Duration (hrs)	Citric acid solubility % P_2O_5	% Conversion of P_2O_5 to citric acid soluble form
Apatite	$Ca(OH)_2$	Quartz				
100	30	10	1000	2·00	3·50	10·29
100	30	20	1000	2·00	4 00	11·76
100	50	20	1100	2·00	4·00	11·76

Table 14 – Effect of temperature on citric solubility of Apatite when calcined with Na_2CO_3 and quartz

RATIO			Temp. °C	Duration (hrs)	Citric acid solubility % P_2O_5	% Conversion of P_2O_5 to citric acid soluble form
Apatite	Quartz	Na_2CO_3				
100	20	30	1000	2·00	9·00	26·47
100	20	30	1100	2·00	12·00	35·29
100	20	30	1200	2·00	15·70	46·17

Table 15 – Effect of duration of treatment on citric solubility of apatite when calcined at 1150°C with Na_2CO_3 and quartz

RATIO			Temp. °C	Duration (hrs)	Citric acid solubility % P_2O_5	% Conversion of P_2O_5 to citric acid soluble form
Apatite	Quartz	Na_2CO_3				
100	10	20	1150	2·00	12·00	35·29
100	10	20	1150	2·50	15·50	45·58
100	10	20	1150	3·00	17·00	50·00

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

From these results it is evident that NaOH and Na_2CO_3 has a better effect on apatite than $Ca(OH)_2$ in rendering it to more citric soluble form. By increasing duration of treatment it is clear that NaOH and Na_2CO_3 would give high citric soluble P_2O_5 content (Figure 4).

But in Sri Lanka since the price of NaOH is high, the use of NaOH is not economical. Hence further experiments were carried out using Na_2CO as a mineralizer.

3·6 Calcination with Na_2CO_3

Table 14 and Figure 3 show that when temperature is increased by keeping Apatite : Na_2CO_3 : Quartz ratio constant, citric acid soluble P_2O_5 % is also increasing. Table 15 and Figure 4 show that reaction comes to near completion when sample is heated for 3 hrs.

The product obtained by this calcination may be of rhenania type PO_4 [12] Hence the possible reaction is

$$Ca_5(PO_4)_3F + 2\,Na_2CO_3 + SiO_2 \longrightarrow 3\,CaNaPO_4 + Ca_2SiO_4 + 2\,CO_2 + NaF$$

By this way P_2O_5 in Eppawela Apatite can be converted to 50 % citric soluble form and can be used as a fertilizer.

Further calcination studies were carried out with Na_2CO_3 alone to find ou the influence of Na_2CO_3 and quartz separately on apatite. The results obtained are as follows (Table 16.).

Table 16 – Effect of duration of treatment on citric acid solubility when apatite is calcined with Na_2CO_3 at 1150°C

RATIO		Temp. °C	Duration (hrs)	Citric acid solubility % P_2O_5	% Conversion of P_2O_5 to citric acid soluble form
Apatite	Na_2CO_3				
100	20	1150	2·00	19·50	57·35
100	20	1150	2·50	21·25	63·23
100	20	1150	3·00	23·00	67·64
100	25	1150	2·00	22·00	64·70
100	25	1150	3·00	26·00	76·47
100	25	1150	4·00	27·60	81·17

By comparing results of Table 16 and 15, it is clear that Na_2CO_3 alone has a better effect on apatite than No_2CO_3 and quartz in beneficiation. Also it shows that 81% of P_2O_5 in apatite can make citric soluble by this type of calcinations.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Further experiments were carried out by varying the temperature of treatment by keeping the molecular ratio and duration of treatment constant (Table 17) and also varying the Apatite : Na_2CO_3 ratio by keeping temperature and duration of treatment constant (Table 18). Results were as follows:

Table 17 – Effect of Temperature of treatment on citric acid solubility of apatite by keeping the material ratio and duration of treatment constant

RATIO		Temp. °C	Duration (hrs)	Citric acid solubility % P_2O_5	% Conversion of P_2O_5 to citric acid soluble form
Apatite	Na_2CO_3				
100	20	900	3·00	12·41	36·50
100	20	1000	3·00	14·31	42·08
100	20	1100	3·00	16·46	48·41
100	20	1150	3·00	23·00	67·64
100	20	1200	3·00	27·35	80·44
100	20	1300	3·00	32·78	96·41

Table 18 – Effect of Apatite : Na_2CO_3 ratio on citric acid solubility by calcining at 1150 °C for 3·0 hrs

RATIO		Temp. oC	Duration (hrs)	Citric acid solubility % P_2O_5	% Conversion of P_2O_5 to citric acid soluble form
Apatite	Na_2CO_3				
100	15	1150	3·00	16·46	36·50
100	20	1150	3·00	23·00	67·64
100	25	1150	3·00	26·00	76·47
100	30	1150	3·00	27·20	80·00
100	35	1150	3·00	26·46	77·87
100	40	1150	3·00	25·77	73·79
100	45	1150	3·00	25·08	73·76
100	50	1150	3·00	22·7	66·76
100	100	1150	3·00	17·15	50·44

The variation of citric acid solubility of Eppawela apatite, when calcined with Na_2CO_3 can be clearly seen in Figures 3, 4 and 5. From X-ray powder analysis studies of these products (Figure 1) and results that had been obtained following conclusions can be achieved.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

a) Increase of temperature of treatment will increase the citric soluble P_2O_5 % considerably when Apatite to Na_2CO_3 ratio was kept constant (Figure 3). X-ray studies (Figure 1) showed the formation of various sodium calcium phosphates[10] (Table 19).

Table 19 – Various phases present in calcined product of apatite with Na_2CO_3 at different temperatures by keeping apatite: Na_2CO_3 – 100 : 20 and for duration of 3 hrs-

Temperature °C	Phases present in the product	Phases present in the residue after dissolving in 2% citric acid
900	$Na_3Ca_6(PO_4)_5$ (ie. 2·4 CaO. 0·6 Na_2O. P_2O_5) Fluorapatite (unreacted)	Fluorapatite
1000	$Na_3Ca_6(PO_4)_5$ + α – Na Ca PO_4 (2 CaO. Na_2O. P_2O_5)	Fluorapatite
1100	$Na_3Ca_6(PO_4)_5$ + α – Na Ca PO_4 + β – Na Ca PO_4	Fluorapatite
1150	— do —	Fluorapatite
1200	β – Na Ca PO_4	Fluorapatite (distorted)
1300	β – Na Ca PO_4	Fluorapatite (distorted)

This means that,

i. At lower temperatures low Na_2O / CaO phosphates are formed by reacting with lesser amount of Na_2O from Na_2CO_3.

ii. At higher temperatures high Na_2O / CaO phosphates are formed with higher amount of Na_2O from Na_2CO_3.

All these phosphates are in citric soluble form. Pure fluorapatite found in the residue may be from unreacted rock and this amount decreased with increasing temperature.

b) Increase in duration of treatment will increase the citric acid solubility considerably until it reaches a treatment duration of 3 – 4 hours. Hence reaction with Na_2CO_3 is complete only after 3 hrs. (Figure 4)

c) Highest citric acid soluble P_2O_5 % can be obtained by using 30 - 40 parts of Na_2CO_3 to that of 100 parts of Apatite (by weight) on calcinations at 1150°C. This is clearly shown in Figure 5. Increase or decrease of

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

$Na_2 CO_3$ ratio at 1150°C lower the citric soluble $P_2 O_5$ percentage in the final product. X-ray studies of the products obtained (Figure 6) showed the presence of various sodium calcium phosphates (eg. $NaCaPO_4$, $Na_3 Ca_6 (PO_4)_5$ etc.). The residue obtained after dissolving in 2% citric acid contained only pure fluorapatite (Figure 7 (A)), if $Na_2 CO_3$ ratio is less than 30 - 35 to 100 parts by weight of apatite.

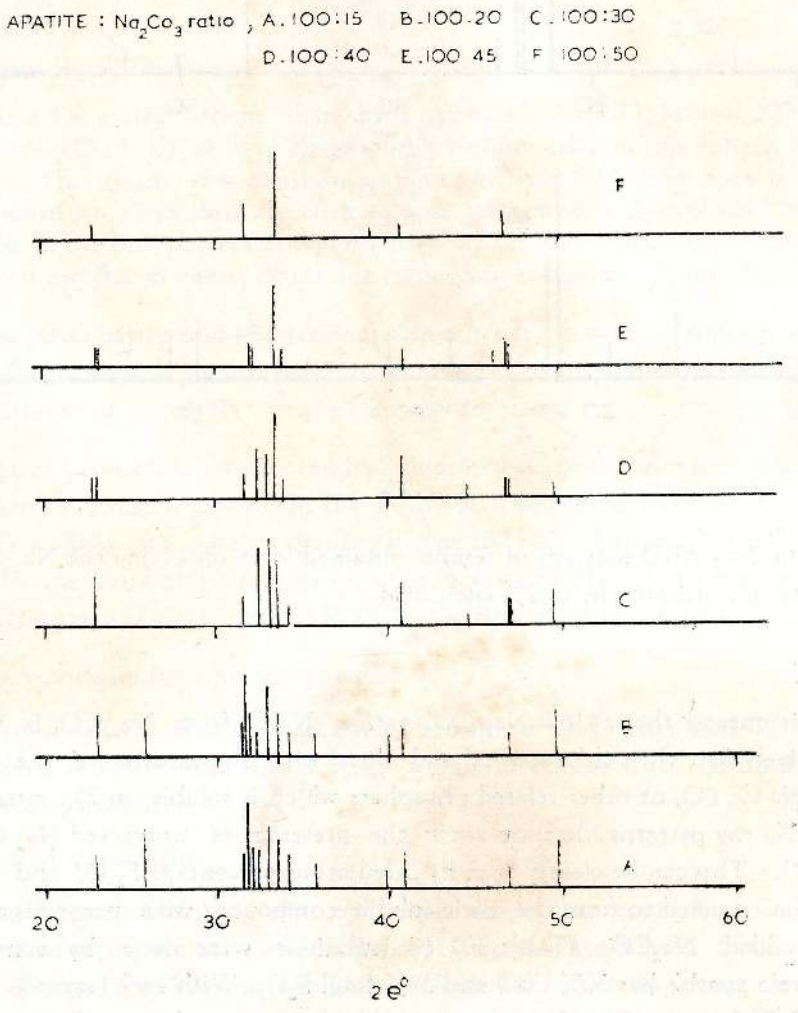


Figure 6 — XRD patterns of products obtained when apatite is calcined at 1150°C for 3·0 hrs with varying amounts of $Na_2 CO_3$.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

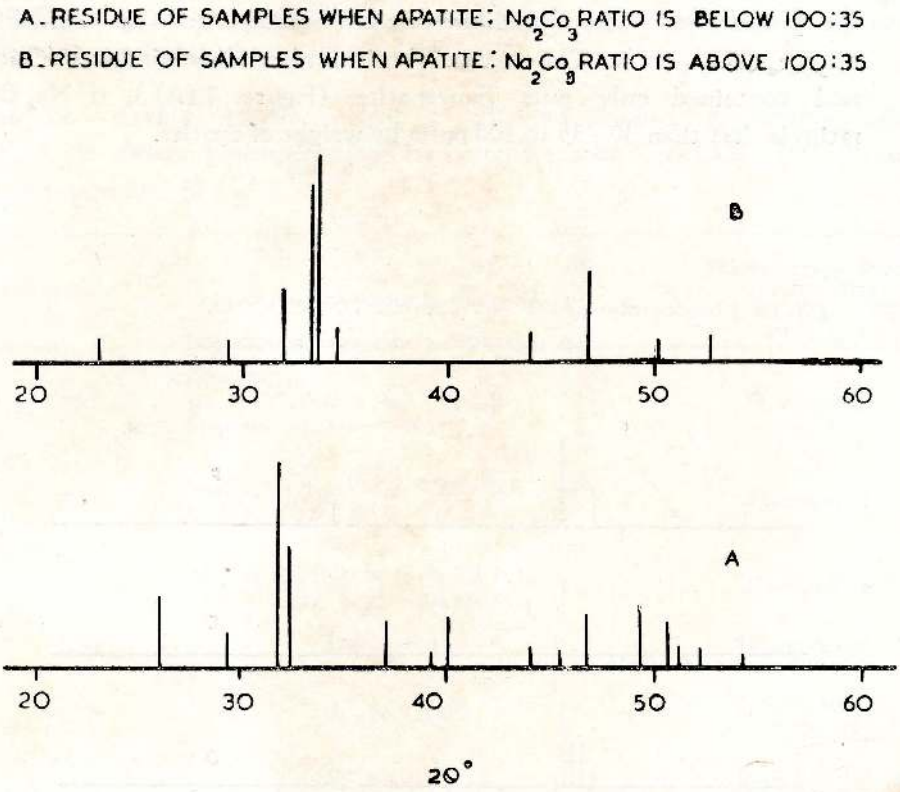


Figure 7 — XRD patterns of residue obtained after dissolving the $Na_2 C0_3$ + Apatite calcined samples in 2% citric acid.

This means that at low $Na_2 CO_3$ ratios, Na_2O from $Na_2 CO_3$ is reacting exclusively first with chlorapatite and then with fluorapatite of the rock to form Na Ca PO_4 or other related phosphate which is soluble in 2% citric acid. (Also X-ray patterns do not show the presence of unreacted $Na_2 CO_3$ or $Na_2 O$). This can be clearly seen by calculating percentage $P_2 O_5$ and percentage CaO reacted to form the citric soluble compound with percentage $Na_2 O$ from added Na_2CO_3. (Table 20) (Calculations were done by considering Eppawela apatite has 50% CaO and 34% total P_2O_5. With each sample percentage CaO that was reacted to give citric soluble compound was calculated from its citric soluble $P_2 O_5$% and $Na_2 O$% was calculated by considering that all Na_2CO_3 was reacting with apatite to give a citric soluble product).

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 20 – Ratio of CaO: Na_2O : P_2O_5 in the citric acid soluble product that had formed with low Na_2CO_3 ratios at 1150 °C for 3 hrs duration.

Reacting Materials		Citric soluble P_2O_5%	Weight Ratio		
Apatite	Na_2CO_3		CaO	Na_2O	P_2O_5
100	15	16·46	1·47	0·530	1
100	20	23·00	1·47	0·50	1
100	25	26·00	1·47	0·56	1
100	30	27·20	1·47	0·594	1

From Table 20 it is clear that until Apatite : Na_2CO_3 ratio is 100 : 30, the CaO : Na_2O : P_2O_5 ratio of citric soluble sodium calcium phosphate, is nearly equal. This means that constant amount CaO and P_2O_5 from rock is reacting with all of Na_2O to form the citric soluble compound. Rest of CaO and P_2O_5 will be in the product as fluorapatite itself. This is evident from X-ray results and cell dimension values of residue fluorapatite obtained (Figure 8).

The XRD patterns of the residual fluorapatite showed a shifting of peaks with the increase of sodium carbonate (Figure 7 (B)). This may be due to the substitution of excess Na^+ in the fluorapatite lattice.

The cell constants of the residual fluorapatite obtained after dissolving in 2% citric solution is plotted in the Figure 8. The cell constants are nearly same upto a sodium carbonate to apatite ratio of 100 : 30. Beyond this cell constant "a" decreases and "b" increases with the increase of Na_2CO_3 percentage. Also the percentage of citric soluble P_2O_5 in the final product decreases.

Colour and the form of the product

The colour of the products varies from brown to grey when Na_2CO_3 ratio is increased at 1150°C and the samples which give high citric solubility had a sintered appearence.

Effect of quenching

It was observed when carrying out experiments that rapid air quenching is needed to get better citric acid solubility in a given condition. Since the product obtained had low water solubility (Table 21) further experiment was carried out with calcined samples by quenching into water. The citric solubility of this sample was very low compared to air quenched sample and X - ray patterns showed the sample had more unreacted fluorapatite.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

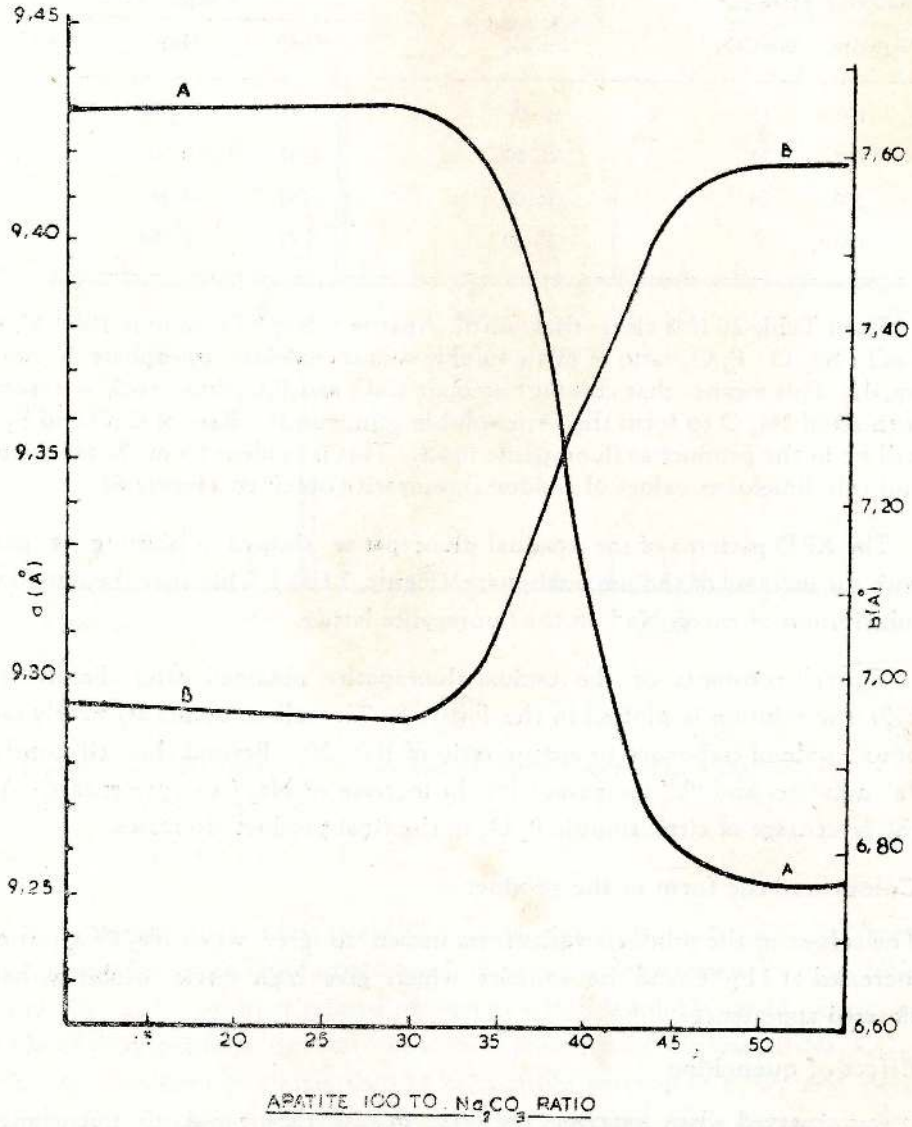


Figure 8 — Variation of cell dimensions of unreacted fluorapatite when raw apatite is calcined with varying amounts of Na_2CO_3 at 1150°C for 3 hours.

A — Variation of cell dimension a

B — Variation of cell dimension b

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 21 – Effect of water quenching and air quenching on citric soluble P_2O_5% on Na_2CO_3 calcined Apatite.

RATIO		Temp. °C	Duration (hrs)	Air Quenching		Water Quenching
Apatite	Na_2CO_3			water sol. P_2O_5%	citric sol. P_2O_5%	citric soluble P_2O_5%
100	20	1150	3·00	3·17	23·00	6·44

Hence it is clear that to obtain high citric soluble P_2O_5 content with calcinations with soda ash at 1150° C, apatite to Soda ash ratio must be in the range of 100 : 30 - 35 and duration of treatment is 3 hrs. The sample should be quenched rapidly in air.

Conclusions

1) Eppawela apatite itself is not suitable as a fertilizer since it has low citric soluble P_2O_5 percentage. (ie. out of 34% P_2O_5 available only 3% is in citric soluble form).

2) It can be converted to a product having more PO_4 in citric soluble form by calcining with sodium carbonate, quartz or paddy hull ash. With sodium carbonate 80% of P_2O_5 can be made citric soluble whereas with quartz and paddy hull ash only 50% can be converted. Calcination of a mixture Apatite + Quartz + Na_2CO_3 will also give 50% conversion.

3) Soda Ash calcination has to be done with controlled amounts of apatite and soda ash to get high citric soluble value and less soda ash consumption. It was found at 1150°C a ratio of Apatite : Na_2CO_3 of 100 : 30—35 at a duration of treatment for 3 hrs, would give best results.

4) If apatite is calcined with controlled amounts of Na_2CO_3 at temperatures above 1300° C, over 95% P_2O_5 in rock can be converted to citric soluble form. For example when a charge consisting of 100 parts of rock apatite, 20 parts of soda ash was calcined at 1300°C for 3 hours a product of 96% citrate soluble P_2O_5 was obtained.

5) There is a possibility to use this product as a phosphate fertilizer for short term crops instead of imported superphosphates, since this type of sodium thermophosphates are presently being used as fertilizers in many countries of the world. But before coming to a firm conclusion one has to carry out field trials with the product.

Acknowledgement

The authors wish to express their gratitude to the Director and the Deputy Director of the C. I. S. I. R. for their encouragement and the staff of the section of Mineral Technology of C. I. S. I R. for their help in preparing this paper.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

References

1 AMARASIRI, S. L., & AMBEPITIYA V. H., (1976) *A new Phosphate Fertilizer product from Eppawela Apatite, paper released from Central Agricultural Research Institute, Peradeniya,*

2 ASSOCIATION OF OFFICIAL ANALYTICAL CHEMISTS, 12th Edition (1975) *Officia Methods of Analysis.*

3 CEYLON FERTILIZER CORPORATION (1974) *Long range fertilizer projections in Sri Lanka.*

4 DEANS, T. *et al,* (1972) *Mineralogy and Chemistry of the rock phosphate deposit at Eppawela. Institute of Geological Sciences (U. K.)*

5 DIAS, H. W., & JAYASEKERA K. S., (1978) *Acidulation studies on Eppawela Apatitte Proceedings of Sri Lanka Association for the Advancement of Science*

6 GREMILLION, R. G., McCLELLAN G. H. & LEHR J. R., (1975) *Characterization of, Phosphate rock from Sri Lanka, Tennessee Valley Authority, Alabama.*

7 HERATH, J. W., (1975) *Mineral Resources of Sri Lanka. Economic Bulletin 2, Geological Survey Department.*

8 JAYASEKERA, K. S., & DIAS H. W., *Study of the breakdown of Eppawela Apatite by sodium carbonate, Proceedings of Sri Lanka Association for the Advancement of Science (1978).*

9 JEFFERY, P. G. *Chemical methods of rock analysis. Pergamon Press, Oxford, (1970)*

10 JOINT COMMITTEE ON POWDER DIFFRACTION STANDARDS. *Powder Diffraction File*

11 SILVA K. K. M. W. *Geology and Nature of Apatite deposit at Eppawela, Department of Geology, Peradeniya.*

12 ZEITSCHRIFT FUR ANGEWANDTE CHEMIE, (1922) *abstracted and translated by Lloyd S. J., in American Fertilizer 57, (13) p 38*

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1)

A Theorem on the Congruences $[\frac{1}{2}(p-1)]! \equiv \pm 1 \pmod{p}$.

M. VELUPPILLAI

Department of Mathematics, University of Peradeniya, Peradeniya, Sri Lanka.

(Date of receipt : 16 May 1979)

(Date of acceptance : 30 January 1981)

1. Introduction.

From Wilson's theorem, we have for any prime $p = 3 \pmod 4$,

$[\frac{1}{2}(p-1)]! \equiv \pm 1 \pmod p$. For some primes the plus sign holds and for some the minus sign hold. Krönecker has derived a rule, mentioned in [1], to decide which of the two signs $\pm$ holds in the above congruence. The object of this paper is to derive a simple formula depending only on the class number $h(-p)$.

2. Theorem:

If p is a prime $\equiv 3 \pmod 4$, then

$[\frac{1}{2}(p-1)]! \equiv -1 \pmod p$, if $h(-p) \equiv 1 \pmod 4$

$\equiv +1 \pmod p$, if $h(-p) \equiv 3 \pmod 4$

3. Proof:

We have $[\frac{1}{2}(p-1)]! \equiv \pm 1 \pmod p$.

Then $\left(\dfrac{[\frac{1}{2}(p-1)]!}{p}\right) \equiv \left(\dfrac{\pm 1}{p}\right) = \pm 1$

Hence $[\frac{1}{2}(p-1)]! \equiv (-1)^n \pmod p$ where n is the number of positive quadratic non-residues of p which are less than or equal to $\frac{1}{2}(p-1)$.

Now, let m be the number of positive quadratic residues of p which are less than or equal to $\frac{1}{2}(p-1)$.

Then $m+n = \frac{1}{2}(p-1)$ (1)

If $p = -1 \pmod 8$, then $h(-p) = m-n$ (2)

and if $p \equiv 3 \pmod 8$,

$h(-p) = \frac{1}{3}(m-n)$. (See e.g [2])

From (1) and (2), we have, if $p \equiv -1 \pmod 8$, then

$$n = \tfrac{1}{4}(p-2h-1)$$

and from (1) and (3), we have, if $p \equiv 3 \pmod 8$, then

$$n = \tfrac{1}{4}(p-6h-1).$$

In both cases n is odd if $h \equiv 1 \pmod 4$ and n is even if $h \equiv 3 \pmod 4$

Hence the theorem.

References

1. AYOUB, R. (1963) *An Introduction to the Analytic Theory of numbers*, Amer. Math. Soc. p. 300—301.

2. USPENSKY, J.V. & HEASLET, M.A. (1939) *Elementary Number Theory*, New York and London p 156—157.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

J. Natn Sci. Coun. Sri Lanka 1981 9 (1) :

Laboratory Investigations on the Repellent and Narcotic Properties of Steam Distillates of Local Plant Extracts to *Sitotroga cerealella* (Olivier)

S. R. KRISHNARAJAH AND V. K. GANESALINGAM

Department of Zoology, University of Jaffna, Jaffna, Sri Lanka.

(Date of receipt : 23 February 1979)

(Date of acceptance : 12 February 1981)

Abstract ; The study was undertaken to determine suitable local plant extracts to be used against *Sitotroga cerealella*, an important pest of paddy in Sri Lanka.

A 'Y' shaped insect olfactometer was used to determine the behaviour of the moths towards the materials used. Additional experiments were conducted in large petridishes to find the toxic effect of the vapor of these materials. Several replicates were taken in each case.

Statistical analysis of the data obtained in the experiments showed that among the materials used, the steam volatile constituents of *Vitex negundo* appeared to be a promising repellent and the vapour of citronella oil was toxic to *Sitotroga cerealella*.

1. Introduction

Effective natural enemies are not available for *Sitotroga cerealella*, an important pest of paddy in Sri Lanka. The use of chemicals to control it may not be advisable due to their residual effect. Although maintenance of a certain standard of cleanliness in storing paddy may reduce infestation[1] it is not possible to reduce infestation appreciably by this method. Therefore, the feasibility of using a suiatable local repellent was investigated in the laboratory.

This study was undertaken to find suitable plant extracts that could be used against the pest in stored paddy. It was thought that the use of local resources as repellents would be of economic value for controlling the pest.

2. Materials and Methods

The moths were reared on paddy under laboratory conditions in bottles (375 c. c.) covered with perforated plastic caps for ventilation. Each newly emerged female was introduced into a bottle containing newly emerged males. Immediately after copulation the female was collected in separate bottles containing paddy sterilised at 80° C for twenty minutes. The 1st generation moths emerged after about a month. One-day-old moths, both females and males in equal number were collected by means of an aspirator and used in these experiments.

A simple 'Y' shaped insect olfactometer similar to that used by Hershberger and Smith[2] was used in this experiment with an additional syringe to push the experimental specimens from the main chamber to the 'Y' tube (Figure 1). A current of air was drawn by a suction pump along both arms of the olfactometer simultaneously and passed out through the outlet of the main stem. The material under investigation was placed in a small bottle, through which air was passed so that its vapour passed into one arm of the olfactometer, while the

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

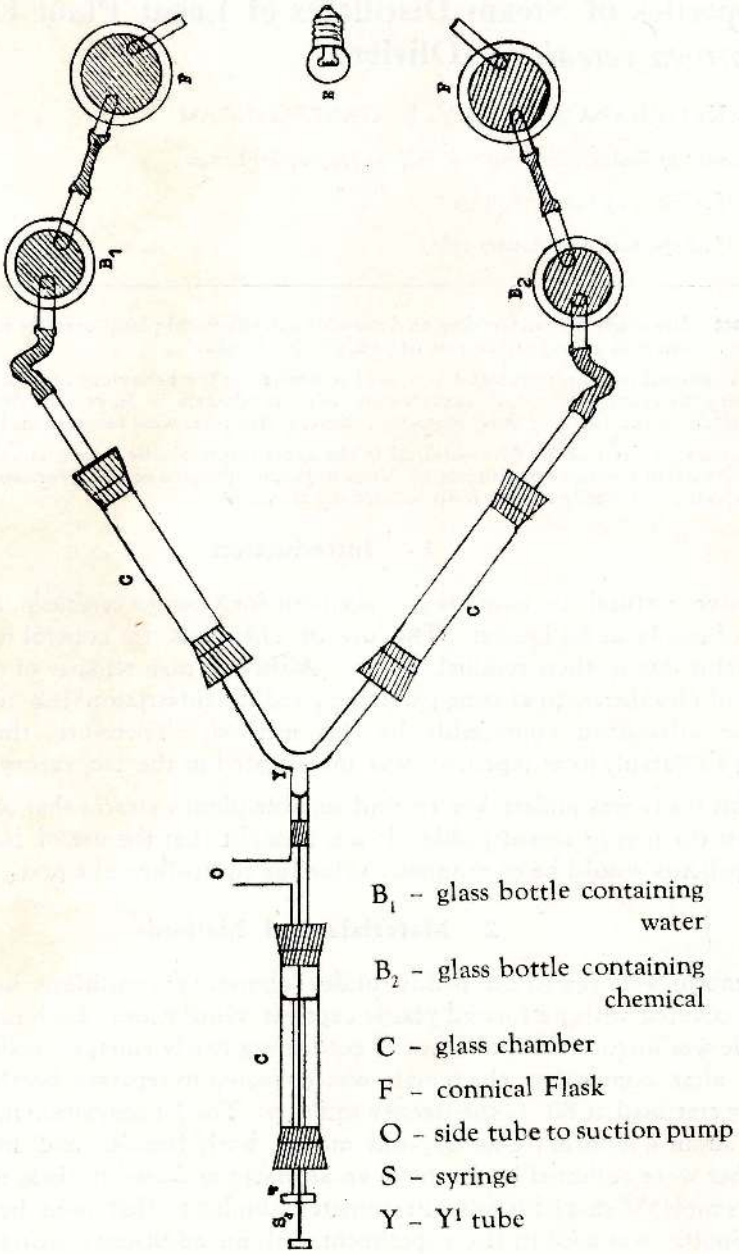


Figure 1 – 'Y' shaped insect olfactometer.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

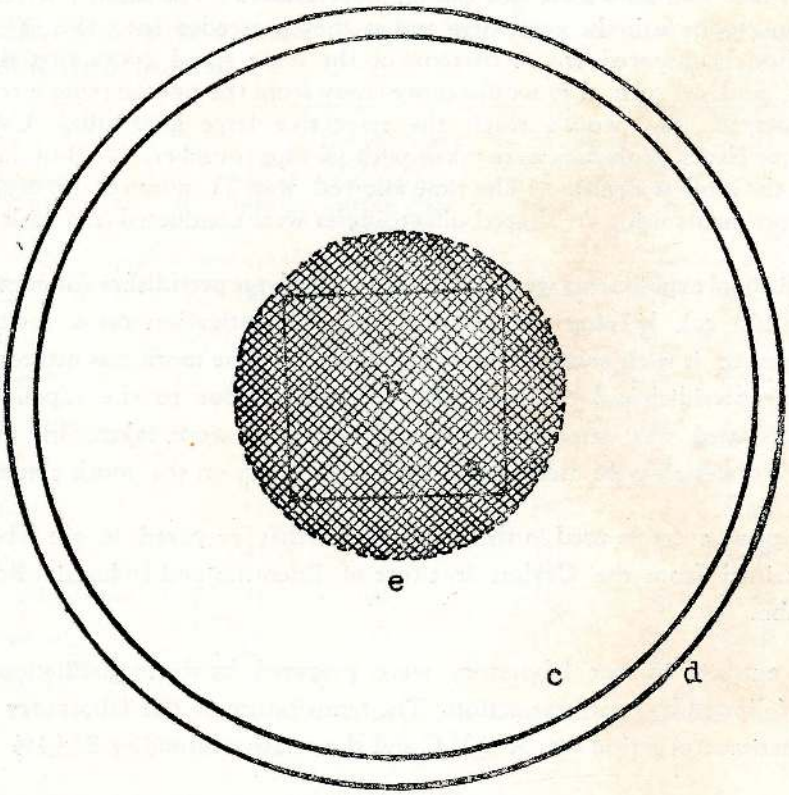


Figure 2 A

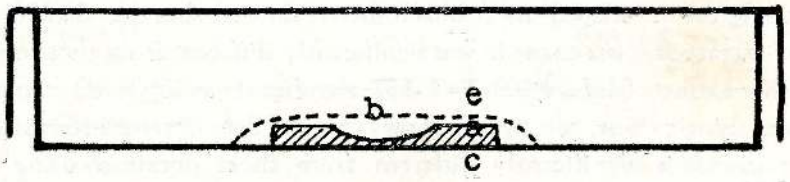


Figure 2 B

Figure – 2 Apparatus used to determine the time taken for the moths to die due to the vapour of the plant extracts.

A. Top view.
B. Side view

a. cavity side
b. plant extract
c. petridish
d. cover
e. mesh

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

other arm carried air which has passed over water. The moths were introduced in groups into the main stem and as they proceeded into the 'Y' shaped tube, they encountered two air currents of the same speed containing different odours, and as such they would move away from the one carrying a repellent. Subsequently, they would reach the respective large glass tubes. Using this technique eleven replicates were taken with various numbers (2-6) of moths in almost the same sex ratio. (The time allowed was 11 minutes in each case). The experiments using 'Y' shaped olfactometer were conducted in a dark room.

Additional experiments were conducted using large petridishes (diameter 9cm; volume 100 cc), keeping the material under investigation on a cavity slide and covering it with another petridish (Figure 2). The moth was introduced into a large petridish and the time taken for it to die due to the vapour of the materials used was determined. Several replicates were taken. In this way different materials were used to find their toxic effect on the moth concerned.

The plant extracts used in this study were either prepared in our laboratory or obtained from the Ceylon Institute of Scientific and Industrial Research, Colombo.

The extracts in our laboratory were prepared by steam distillation of the leaves, followed by other extraction. The temperature of the laboratory during the experimental period was $30° \pm 2°$ C and the relative humidity $80 \pm 4\%$

3. Results

Statistical analysis using analysis of variance[3] for the data obtained in the experiments using the 'Y' shaped insect olfactometer, showed that the data obtained with the extract of *Vitex negundo* was significantly different from those obtained using other extracts (d. f = 9/100, F = 1·682 significant at 10% level). Analysing the results using t - test, pair by pair shows that the data obtained for the extract of *Vitex negundo* is significantly different from those obtained using other extracts. The "Chi" square test applied to the data obtained using the extract of *Vitex negundo* showed that there is a significant difference between the number of moths moving into the arm ($X^2 = 12 \cdot 736$, $P < 0 \cdot 005$). The number moving into the arm containing the extract of *Vitex negundo* was significantly less than the number moving into the other arm - (Table 1).

With regard to the time taken for the moths to die due to vapour toxicity of the materials used, it was found that among the materials used, Citronella oil vapour, killed within the shortest time (14 minutes an average of 11 readings taken in each case - Table 2).

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 1 – The direction of movements of *Sitotroga cerealella* in the 'Y' shaped olfactometer when given a choice between the plant extracts (oil) and water. (* significantly different).

Materials	Total No. of Insects	No. of insects moved into the arm containing plant extract	No. of insects moved into the arm containing water	Non-response	X^2	P
Citronella oil	50	12	28	10	6·4	*<0·025
Clove oil	53	17	14	22	0·029	<0·900
Margosa oil	56	20	22	14	0·095	<0·900
Cinnamon leaf oil	47	13	21	13	1·82	<0·250
Eucalyptus oil	46	9	18	19	3·00	<0·050
Lime leaf oil	42	21	15	6	0·111	<0·750
Vitex oil	43	8	30	5	12·736	*<0·005
Camphorated margosa oil	54	18	31	5	3·447	<0·050
Lemon grass oil	64	13	13	38	0·000	<0·095
Ocimum extract	39	10	16	13	1·384	<0·250

Table 2 – The time taken for the moths to die due to the toxic effect of the vapour of the materials used. (Average of 11 readings in each case).

Materials	Average time taken for death (mins.)
Citronella oil ..	14
Clove oil ..	16·5
Vitex oil ..	18
Cinnamon leaf oil ..	18
Lemon grass oil ..	19
Margosa oil ..	>30
Eucalyptus oil ..	>30
Lime leaf oil ..	>30
Camphorated Margosa oil ..	>30
Ocimum extract ..	>30

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

4. Discussion

The study revealed that of all the plant extracts tested for potent repellency against *Sitotroga cerealella*, the extract of *Vitex negundo* could be considered as a promising repellent. The other materials were unsuitable for this purpose. The odour of the extract of *Vitex negundo* seems to be intolerable, as it is to some animals and to man. It is probable that the olfactory organs of the moth are extra sensitive to the odour of the extract of *Vitex negundo*.

From the present study one could conclude that the oil of Citronella (*Andropogon nardus* – Lenabatu type) is the most toxic material used in these experiments, as it takes the shortest time to kill the pest. Although Citronella (Lenabatu type) seems to have a repellent property to mosquito species as reviewed by Wijesekera,[5] how it causes death in *Sitotroga cerealella* is not known.

The extract of Black pepper, *Piper nigrum* (L) has been used against rice weevil, *Sitophilus oryzae* (L), and cowpea weevil, *Callosobruchus maculatus* (F).[4] In similar manner, *Vitex negundo* could be made use of as repellent and poison against *Sitotroga cerealella*.

The toxicity of Black pepper was not attributed to the presence of piperine alone. Several other chemical components of Black pepper may be ascribed for its toxicity in conjunction or in synergism with piperine.[4] Likewise, *Andropogon nardus* contains about 47 constituents[5] but the components with toxic or repellent qualities have not been identified. In the case of *Vitex negundo*, even the chemical consitituents have not been investigated.

Further investigations on the chemical constituents and their effects individually, or in conjunction with others, so as to determine the most promising composition, for repellency or toxicity are necessary.

Acknowledgements

We thank Prof J. B. Selliah for his assistance in the statistical analysis and the staff of the Department of Zoology, University of Jaffna, for their assistance in the experiments. We also gratefully acknowledge the assistance of the staff of the Ceylon Institute of Scientific and Industrial Research in providing us with the plant extracts. This was supported by a grant from the National Science Council of Sri Lanka, which is gratefully acknowledged.

References

1 GANESALINGAM, V. K. (1975) – *Insects in paddy and rice grain storage in Kandy district.* Proc. Cey. Assn. Advmt. Sci. **1** : 45 – 46.

2 HERSHBERGER, W. A. & SMITH, M. P. – (1957) – *Conditioning in Drosophila melanogaster.* Anim. Behav. **15** : 259 – 626.

3 SNEDECOR, W. G. (1952) – *Statistical Methods, The Iowa State University Press, Ames Iowa U. S. A. 534 p.*

4 SU, H. C. F. (1977) – *Insecticidal properties of black pepper to rice weevils and cowpea weevils.* J. Econ, Entomol **70** : 18 – 21.

5 WIJESEKERA, R. O. B. (1973) – *The Chemical Composition and analysis of Citronella oil,* J. Natn. Sci. Coun. Sri Lanka. **2** : 67 - 81.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1):

Nitrogen Use and Economics of Intercropping in Sri Lanka

H. P. M. GUNASENA

Faculty of Agriculture, University of Peradeniy, Peradeniya, Sri Lanka.

(Date of receipt : 01 August 1978)

(Date of acceptance : 09 March 1981)

Abstract Field studies to compare N utilization and economics of some intercropped and mono cropped systems were conducted in Sri Lanka using maize / cowpea maize / soybean and maize /mungbean crop combinations. N levels used were 0%, 50% and 100% of the locally recommended rates for the non-legume crop. At all locations, the non legume crop showed a positive response to N under both cropping systems. Yield of the intercropped legume was depressed at higher N levels possibly due to shading effect of the non-legume crop. Total output and gross returns increased with intercropping. The net protein utilization value of the inter cropped system was also higher highlighting the importance of such systems for the subsistence level farmers of developing countries.

1. Introduction

In many Asian and Pacific countries, farmers with limited resources have traditionally multiple cropped their lands to minimize risks associated with growing a single crop, and to assure a more stable subsistence in terms of food, nutrition, and possible income. Using the available family labour and limited capital resources, they have attempted to maximize returns from these cropping systems, generally under low levels of technology. In the developed countries, however, multiple cropping has been viewed as a practice not well suited to modern agriculture, and for this reason, research in this field has been largely inadequate. One of the main reasons for this neglect seems to be that most agricultural research reported in literature has been carried out in countries where monocultures prevail. Since agriculture in these countries is mechanized, research has focused on agricultural systems that have mechanization as one of their major components[2] Due to recent increases in population densities and in prices of agricultural resources, an interest is now developing to seek technological alternatives for increasing food production. Intercropping as a means of optimizing the use of land and other inputs, is starting to receive increased attention.

One of the areas requiring an enhanced understanding in multiple cropping systems is the use of fertilizer nitrogen, a resource which is expensive and not easily available to farmers in developing countries. The present study was therefore designed to evaluate N utilization of intercropped systems, and to compare its economics with corresponding monocropped systems.

2. Materials and Methods

A non-leguminous monocrop system at three N levels was compared with an intercropped system consisting of the same crop inter-planted with a grain legume, at the same levels of N. The study was conducted at Peradeniya and

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Kundasale in Sri Lanka with two experiments at each location. The levels of N selected were 0%, 50% and 100% of 80 kg N/ha, the recommended N for the row crop[1] maize *Zea mays* L. All plots received 100% of the recommended rates of P *(50 kg P/ha)* and K *(65 kg K/ha)* for the row crop only and other cultural practices for both crops uniformly as per local recommendation. In Peradeniya experiment (B) where soybean was used as the legume, it was inoculated with a commercial inoculum, 'Nitrogin S' at the rate of 5 g/kg of seed. Grain legumes used were soybean *Glycine max* (L) Merr. (1 site), cowpea *Vigna unguiculata* L. (1 site) and mungbean *Vigna radiata* L. (2 sites.) Both row and legume crops were planted at the same time. The legume was planted centrally between row crop rows. Treatments were arranged in randomized blocks replicated four itmes. Locations, soil characteristics and other experimental details are indicated in Table 1.

Table 1 – Locations, soil characteristics and other experimental details

Location	PERADENIYA		KUNDASALE	
	(A)	(B)	(C)	(D)
Altitude ..	450 M	450 M	420 M	420 M
Texture ..	clay loam	clay	loamy sand	loamy sand
pH ..	5.3	6.1	6.4	6.4
Organic carbon (%) ..	1.55	1.98	1.04	1.6
Total N (%) ..	0.27	0.1	0.09	0.28
CEC (me/100g) ..	40.8	43.8	4.60	3.96
Available P (me/100g) ..	52.7	15	23.0	52.7
Available K (me/100g) ..	0.54	2.11	0.35	0.64
Available Ca (me/100g) ..	21.94	25.81	3.35	4.00
Available Mg (me/100g) ..	13.60	12.90	0.83	0.76
Available Na (me/100g) ..	0.92	0.93	0.06	0.04
Row crop variety (maize) ..	Thai Composite	Thai Composite	T 48	T 48
Legume ..	cowpea	soybean	mungbean	mungbean
Legume variety ..	Bushitavo	PB. 1	MI. 1	MI. 1
Date of Planting (1976) ..	May 16	July 16	May 20	July 2
Date of harvesting (1976)				
row crop ..	Sep. 20	Nov. 20	Sep. 23	Nov. 8
legume ..	Aug. 30	Oct. 24	Aug. 16	Oct. 10
Spacing, cm				
row crop ..	75 x 25	75 x 25	75 x 75	75 x 25
legume ..	75 x 20	75 x 5	75 x 20	75 x 20
Rainfall during experimental period, mm+ ..	712	721	413	200

+ Supplemental irrigation provided in all cases, as needed

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 2 – Mean yields of row and legume crops under various levels of nitrogen, kg/ha

	Rate of recommended N as %	PERADENIYA						KUNDASALE					
		(A)			(B)			(C)			(D)		
		maize	cowpea	total	maize	soybean	total	maize	mungbean	total	maize	mungbean	total
Intercrop System	100	9·85a	0·48	10·33	6·08ab	1·06	7·14	4·88a	0·08	4·96	1·27b	0·13	1·40
	50	9·53a	0·33	9·86	5·10bed	1·08	6·18	3·86b	0·29	4·15	1·00b	0·15	1·15
	0	7·88b	0·50	8·38	4·25d	1·44	5·69	2·33a	0·43	2·76	0·60c	0·24	0·84
Monocrop System	100	10·16a	—	10·16	6·70a	—	6·70	4·84a	—	4·84	1·67a	—	1·67
	50	10·06a	—	10·06	6·13ab	—	6·13	4·35ab	—	4·35	1·11b	—	1·11
	0	7·88b	—	7·88	5·52ab	—	5·52	3·06c	—	3·06	0·60c	—	0·60

Means in the same column followed by the same letter (s) are not significant at the 5% level (Bayes LSD)

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

3. Results and Discussion

The mean yield of row and legume crops for various locations is given in Table 2.

Yield of row crop: (i) *Effect of* N – Whether intercropped or not, maize showed a positive response to N application, The yield difference between the 50% N and 100% N levels however, was significant in two cases: intercrop in Kundasale (C) and monocrop in Kundasale (D). These results would perhaps suggest a re-examination of what constitutes a "recommended" fertilizer application by farmers.

An interesting feature is that the magnitude of yield response and its trend varied with variety and location. Thus a considerable variation in the yield of maize was recorded among the four sites. The yield ranged from 0·60 – 1·27 t/ha in Kundasale (D) to 7·88 – 9·85 t/ha in Peradeniya, (A). Maize yields in Kundasale trials are too low to make valid comparisons between locations.

ii. *Effect of Cropping System* — As a general trend, maize yields were somewhat depressed by intercropping, particularly at lower N levels. The differences however, were statistically significant only in a few instances: the 0% N level in Kundasale (C) and Peradeniya (B), and the 100% N in Kundasale (D). The yield gap between monocropped and intercropped maize tended to decrease with increasing N levels, except in Kundasale (D). This would indicate that at lower N levels, there is competition between maize and interplanted legumes for N, and that the competitive effect decreases as the N level increases. Agboola and Fayemi[1] studied the effect of intercropping maize with cowpea and mungbean at varying N levels and found that maize yield was not decreased by intercropping at higher N levels. Similar results have been reported by Haizel[6] when maize was intercropped with cowpea in two cropping seasons. Beets[2] concludes that maize could withstand intercropping with compatible crops. These arguments do not help to explain the yield differences in the Kundasale (D) however, possibility of some, as yet unexplained, interaction between maize and mungbean cannot be ruled out. The significant maize yield reduction due to intercropping at 0% N level in Kundasale (C) and Peradeniya (B) may be a reflection of the inability of maize to compete with either mungbean or soybean for the available soil N. Similar conclusions are also reported by Harwood and Banta.[8]

Yield of Legume: Table 2 indicates that, in all cases, the yield of intercropped legume decreased with increasing levels of N application, except in Peradeniya (A). Thus, at all four sites, the 0% N level gave the highest legume yield, regardless of whether it was mungbean, cowpea or soybean, and this tended to decrease with increasing N levels. Shading and competition effects of the row

---

The term "row crop" wherever used will denote the non-leguminous crop being tested; "legume" will designate the inter-planted grain legume.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

crop at higher N levels would probably account for this. Syariffuddin *et al*[14] reported an increase in shading effect and a corresponding reduction in legume yield at higher densities of maize. This suggests that wider spacings of the row crop would be required if higher yields of inter-planted legume is desired.

Yield of total system : The total yield of the intercropped system was higher than that of its monocrop counterpart at all N levels, except in a few cases (Table 2). This suggests that there was better utilization of land, N, and other inputs under intercropping. The magnitude of the difference, however, varied with crop combinations and location. Results of these and some other studies so far reported indicate that a new potential for total production systems thus exists if plant types are selected to exploit soil and climates while maintaining optimum plant populations and other cultural practices (Jeganathan *et al.*,[10], and Pinchinat and Oelsligle,[12], for maize – soybean combinations; Roquib and Kundu[13], for maize – groundnut and rice-mungbean combinations; and Beets[12], for rice-soybean and maize-soybean combinations). On the other hand, Koli[11] and IRRI[9] report a reduction in yield of both crops in intercropped systems. One of the deficiencies of results so far obtained is that most research concerned with inter- cropping has been conducted with crop varieties selected for performance in pure culture. As Harper[7] points out, varieties that are expected to perform better in mixtures should be specifically bred for this purpose. Data reported by Digkstra and De Vos[4] who tested clover selections in both pure and mixed stands with pasture grass support this view.

Human nutritional value. The effect of cropping systems and N level on the crude protein content of maize and cowpea in Peradeniya (A), although not subject to statistical analysis, shows that, while crude protein of cowpea was not affected by N levels, that of maize increased in both monocrop and intercrop systems (Table 3). At rates below the 100% N level, crude protein content of maize under monocrop was slightly higher than under intercrop; at the 100% N level, on the other hand, crude protein content was higher under the intercrop system the combined maize-cowpea crude protein content of the intercrop system was much higher than that of the maize monocrop system at all N levels. From a practical human nutrition standpoint, the significant points to note are:

Table 3 – Effect of cropping systems and N level on crude protein content of maize and cowpea, Peradeniya (A)

Level of N, kg/ha	Crude Protein Content, %		
	Monocrop System	Intercrop System	
	maize	maize	cowpea
80	10·94	11·70	24·90
40	9·93	8·96	24·90
0	8·88	8·35	25·00

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

1. The maize- cowpea intercrop yielded approximately 10% more crude protein/ ha, compared to maize monocrop, at the same N levels.
2. Since cowpea has a higher lysine content (427 mg/g *vs* 167 mg/g N in maize), the concurrent use of cowpea and maize as food should have much better net protein utilization than maize alone.[5]

The data is inadequate to make further explanations, and indicates that there is a need for much further research on the human nutrition aspects of intercropping.

Economics. The total economic value of the produce obtained (t/ha) was based on international wholesale crop prices in some cases and the local market prices prevailing at the time of study in other cases (Table 4). The differentially higher cost incurred in the intercrop for family labour has not been taken into account, since family labour is readily available in most developing countries.

Table 4 – Economics of mono and intercrop systems+

	Gross returns at various N levels, Rs/ha					
	Zero N/		40 kg N/ha		80 kg N/ha	
	monocrop	intercrop	monocrop	intercrop	monocrop	intercrop
Peradeniya (A)						
maize	3,325·0	3,350·2	4,275·6	4,050·2	4,316·2	4,191·6
cowpea	—	432·0	—	274·4	—	417·2
TOTAL	3,325·0	3,782·2	4,275·6	4,324·6	4,316·2	4,608·8
Peradeniya (B)						
maize	3,175·2	1,806·0	2,608·2	2,167·2	2,850·4	2,583·0
soybean	—	1,741·6	—	1,300·6	—	1,267·7
TOTAL	3,175·2	3,547·6	2,608·2	3,467·8	2,850·4	3,850·0
Kundasale (C)						
maize	1,299·2	991 2	1,841·0	1,642·2	2,063·6	2·074·8
mungbean	—	242·2	—	366·8	—	50·4
TOTAL	1,299·2	1,233·4	1,841·0	2,009·0	2,063·6	2,125·2
Kundasale (D)						
maize	266·0	266·0	466·2	425·6	708·4	541·8
mungbean	—	1,275·4	—	733·6	—	609·0
TOTAL	266·0	1,541·4	466·2	1,159·2	708·4	1,150·8

+ Average wholesale international price used for maize, cowpea and soybean for mungbean the wholesale price prevailing in Sri Lanka at the time of study has been used due to non-availability of data on international prices.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

In all four experiments, regardless of crop combinations used, the intercrop system provided higher returns than the monocrop system at corresponding N levels, the increase varying from a non-significant 1% in Peradeniya (A) (50% N level) to a highly significant 416% in Kundasale (B) (0% N level). The only exception where the monocrop yielded more returns was the 0% N level in Kundasale (C). The difference amounts to 5%. Intercropping with a higher value crop such as mungbean provides for a higher differential return than if a lower value crop such as cowpea is used. Roquib *et al*[13] also found combinations of maize and soybean, rice and soybean, and maize and groundnut to give better monetary returns per unit area; Jeganathan *et al*[10] reported more net profit in intercropped maize and soybean. Beets[2], in a comprehensive study on mono- and intercropping economics in Thailand reported that soybean - maize intercrop could yield 30% more income compared to monocrop maize.

Apart from increased returns, intercrop systems could also provide for a greater protection against risks due to pests and adverse climatic conditions, as these tend to affect various crops differently. A farmer would therefore be able to harvest one crop even if the other crop was completely destroyed.

4. Conclusion

Intercropping with compatible crop combinations increases total output per unit area and therefore provides for more efficient utilization of N, land and other inputs. Depending upon market prices of crops used, total returns to farmers are also increased. Intercropping with legumes appears to enhance the net protein utilization value of the system, highlighting the importance of such management practices for subsistence level farmers of developing countries.

Acknowledgement

Financial assistance received for this investigation from the National Science Council of Sri Lanka is gratefully acknowledged.

References

1 AGBOOLA, A.A. & FAYEMI, A.A. (1970). *African J. Biol. Appl Chem.* **13** (2): 31
2 BEETS, W. C. (1975). *Agric. Env.* 2 (3): 219
3 BEETS, W. C. (1977). *World Crops.* **29** (1): 25
4 DIGJKSTRA, J. & De VES, A.L.F. (1972) *Euphytica.* **21** : 432.
5 FAO (1970). FAO *nutritional studies No.* 24. *Food and Agricultural Organization of the United Nations. Rome,* 38 *&* 52
6 HAIZEL, K.A. *(1974) Ghana J. Agric. Sci.* **7** (3): 169.
7 HARPER, J. L. (1967). *J. Ecol.* **55**: 247
8 HARWOOD, R. R. & BANTA, G.R. *(1974)* Agron. Abstr. 45.
9 IRRI (1975). *International Rice Research Institute, Annual Rept.* 365
10 JEGANATHAN, N.T., MORACHAN, Y.B. & RAMIAH, S. (1974). *Madras agric. J.* **61** (8): 386.
11 KOLI, S. E. (1975) *Ghana J. Agric Sci.* 8 (1): 23
12 PINCHINAT, A. M. & OELSLIGLE, D. D. (1975). Agron: Abstr. 46.
13 ROQUIB, A., KUNDU, A.L. & CHATTERJEE, B.N. (1973). *Indian J. agris Sci* **43** (8): 792.
14 SYARIFFUDDIN, A., EFFENDY S., SMAIL, I. G. & MCINTOSH, J. L. (1974) *Rept. Res: Inst, Agric, Bogar Indonesia.* **12**: 13

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1):

SHORT COMMUNICATION

Hormonal Induction of Lactation in Buffaloes (*Bubalus bubalis*) A Preliminary Study

R. RAJAMAHENDRN, K. P. M. PATHIRANA AND M. THAMOTHARAM*

Department of Animal Husbandry, University of Peradeniya, Peradeniya, Sri Lanka.

(Date of receipt : 23 July 1980)

(Date of acceptance : 1 January 1981)

Development of a reliable procedure to induce lactation in cattle and buffaloes will have considerable practical as well as theoretical significance. Through this procedure, it may be possible to meet the consumer demand of liquid milk, derive profits from uneconomical cows and heifers and salvage unproductive cows that are otherwise culled due to reproductive disorders or prolonged calving intervals.

A review of the literature on artificial induction of lactation reveals that many different procedures have been used to induce lactation in cattle, with exogenous hormones. Generally lactation milk yields were higher following prolonged treatments with various combination of oestrogen and progesterone[2,3,5] as compared to oestrogen alone.[3] Smith and Schanbacher[4] modified earlier procedures by increasing the daily oestrogen dose 100 fold and injecting subcutaneously oestradiol 17 β (0.05 mg/kg bodyweight) and progesterone (0.125 mg/kg bodyweight) dissolved in absolute ethanol, 15 times at 12 hr interval. Although lactogenesis occurred in a majority of animals within 1 to 4 weeks after treatment, rates of milk production varied widely among individuals. However this method has several advantages such as short duration of treatment and also the post treatment oestrus is short and fertile.[1]

The use of above hormone treatment schedule for the induction of lactation in buffaloes has hitherto not been investigated. The purpose of this preliminary study was to determine whether the short term progesterone oestrogen treatment was as effective in inducing lactation in buffaloes as in cattle.

This study was conducted at the National Livestock Development Board Farm, Malsiripura, in the Kurunegala district. Surti buffalo breeds are maintained in this farm. Nine animals, four non-pregnant primiparous cows and five cycling non-pregnant heifers were made available for this study. The

* National Livestock Development Board, Malsiripura.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

average age and weight of cows was 50 months and 400 kg and the heifers 24 months and 300 kg. All the experimental animals were rectally palpated before treatment to assess their ovarian and uterine status and they were allocated at random to two treatment groups. Two cows and two heifers were assigned to Group I and the remaining two cows three heifers were assigned to Group II. Hormones used were oestradiol 17 β (E_2) and progesterone (P) dissolved in absolute alcohol. Hormone injections were given subcutaneously behind the scapula at 12 hr intervals and each dose contained 0.05 mg of E_2 and 0.125 mg of P per kg of body weight. Animals in Group I received 15 injections (7 days) while Group II animals received 29 injections (14 days). An intramuscular injection of dexamethasone sodium phosphate (0.03 mg/kg bodyweight) was given to each animal seven days after the last P+E_2 treatment. Manual stimulation of the mammary glands, twice daily were carried out after the dexamethasone treatment and regular milking was commenced when the glands became full and turgid.

The following observations were made on the animals a) udder and teat development during treatment and lactation, b) the milk yield for 2 weeks after induction, c) the composition of induced milk during peak lactation and d) interval to oestrus and pregnancy following treatment.

Udder and teat developmont were assessed by visual observations and by handling the mammary glands and teats before the first P + E_2 injection and on days 10, 20 and 30 after the first injection. The degree of udder and teat development were more pronounced in heifers than in cows. Maximum development was observed around 30 days after the commencement of P + E_2 treatment. Since pregnancy period in buffaloes is about 310 days, we rationalised that a longer hormone injection period might prove beneficial to mammary development. However no difference was observed in the udder and teat development between the two treatments groups. Lactation was initiated in all the treated animals. However the amount of milk yield as expected was more in cows than in heifers. (Table 1). Of the 4 cows treated 2 of them had peak lactation of about 3 litres. This yield was 75% of their previous lactation yields. Three out of 5 heifers had peak lactation of about 1 litre. The peak lactation yield was attained around 50 to 70 days after initiation of lactation. The composition of milk namely fat%, total solids and solids not fat obtained during the peak lactation were not different from normal milk. The results also indicates that the reproductive state of the animal before the commencement of the treatment did not have any effect on initiation of lactation. Oestrus

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 1 – Average milk yield per day in liters for eleven weeks following hormone induced lactation in Buffalo cows and heifers

TREATMENT Group I	Animal No.	1*	2	3	4	5	6	7	8	9	10	11
Progesterone + Oestradiol 17β for 7 days	62 cows	0·6	1·1	1·7	2·0	2·7	2·7	3·1	3·2	3·4	3·6	3·1
	130 cows	0·004	0·001	—	—	—	—	—	—	—	—	—
(15 injections at 12 hrs interval)	165 heifers	0·02	0·02	0·07	0·09	0·1	0·07	0·05	—	—	—	—
	176 heifers	0·004	0·005	0·2	0·7	1·1	0·9	1·1	0·9	1·0	1·0	0·7
Group II												
Progesterone + Oestradiol 17β for 4 days	163 cows	0·018	0·1	0·3	0·8	0·9	0·8	1·0	0·9	0·8	0·9	0·9
	166 cows	0·028	1·6	2·4	2·6	3·2	3·4	3·6	3·2	3·4	2·9	2·8
(29 injections at 12 hrs interval)	77	0·016	0·5	1·0	0·9	1·3	0·7	0·8	0·8	1·2	0·9	1·1
	78 heifers	0 ·005	0·4	0·8	0·8	0·9	0·8	0·8	0·7	1·1	0·9	1·2
	164	·101	0·4	0·3	0·3	0·2	—	—	—	—	—	—

* Week

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

signs were not exhibited by the animals during the course of the treatment. Most of the animals returned to oestrus within 30 days after treatment and 3 animals were diagnosed pregnant 90 days later.

The results obtained from this preliminary study is very encouraging. Further studies on a large number of reproductive problems of animals should be carried out to determine the lactation length and total milk yield; hormone levels in milk following induction; fertility following induction treatment; cost benefits of this induction treatment, before recommending this procedure to the dairy farmer.

Acknowledgements

The authors wish to acknowledge the cooperation of the Project Manager and staff of National Livestock development Board, Malsiripura, Sri Lanka.

References

1. ERB, R.E., MONK, E.L., MOLLETT, T.A., MALERN, P. & CALLAHAN, C. J., (1976) *J. Anim. Sci.*, 42(3): 644.
2. HANCOCK, J., BRUMPY, P. J. & TURNER, C.W. (1954) *N. Zealand Jl. Sci. Tech nol.* 36 :111.
3. MEITES, J. (1961). *In* S. Kon and A. T. Cowis *(Ed.)* Milk: *The mammary gland and its Secretion*, Vol.IIP.321. Academic Press New York. N. Y.
4. SMITH, K.L. S. & SCHANBACHER, F.L. (1973) *J. Dairy Sci* 56: 738
5. TURNER, C. W., YAMAMOTO, H. & RUPPER J.H.L (1956) *J. Dairy Sci.*, 39: 1717

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1)

SHORT COMMUNICATION

The Influence of Frequency of Feeding on Rumen Concentration of Volatile Fatty Acid (VFA) and Ammonia in Growing Buffalo Calves.

M. C. N. JAYASURIYA*, U. R. MEHRA AND R. S. DASS

Division of Animal Nutrition, Indian Veterinary Research Institute Izatnagar, (U.P.) 243122. India

(Date of receipt : 22 February 1980)

(Date of acceptance : 02 January 1981)

A major criterion for the isotope dilution techniques is that the animal remains in a relatively steady state throughout the experimental period. Unless the animal has reached the steady state, production rates of Volatile Fatty Acid (VFA) or microbial protein cannot be accurately determined. A steady state further makes it possible to extrapolate results on a daily basis.[1]

The present study was conducted to determine how frequency of feeding influences steady state in the buffalo calf with regard to rumen concentration of volatile fatty acids and ammonia.

Four growing male buffalo calves, approximately 3 years of age, fitted with rumen cannulae were used in the study. Animals were randomly divided into two groups of two each. Chaffed green maize was offered *ad libitum* to all animals. After a pre-experimental period of four weeks animals in group 1 were given green maize twice daily at 08.00 and 16.00 hours. Animals in group 2 were fed their daily ration in twelve equal amounts at two hourly intervals. All animals had access to *ad libitum* water.

After a period of three weeks on this feeding regime samples of rumen liquor were drawn from four different sites in the rumen at various time intervals up to 10 to 14 hours using specially built probes covered with fine nylon gauze as described previously.[1] About 20 ml of rumen fluid received in cold Macartney bottles containing 0·2 ml of 10 N sulphuric acid were taken to the laboratory and analysed immediately for total VFA and rumen ammonia by Markham distillation.[4] Rumen liquor samples were further fractionated by gas liquid chromatography (Shimadzu model GC-4 (B) BTF) using a column of chromosorb W-HMDS with tween 80.[2] Nitrogen was used as the carrier gas on the flame ionization detector.

* Present Address: Department of Animal Husbandry, University, of Peradeniya, Peradeniya, Sri Lanka.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Total VFA produced at different time intervals by animals on the two treatments are presented in Figure 1. The concentration of total VFA remained within very narrow limits at different times as a result of two hourly feeding. Similar results have been reported earlier when Know and Ward[3] compared twice a day feeding with eight times feeding using brown Swiss animals on a

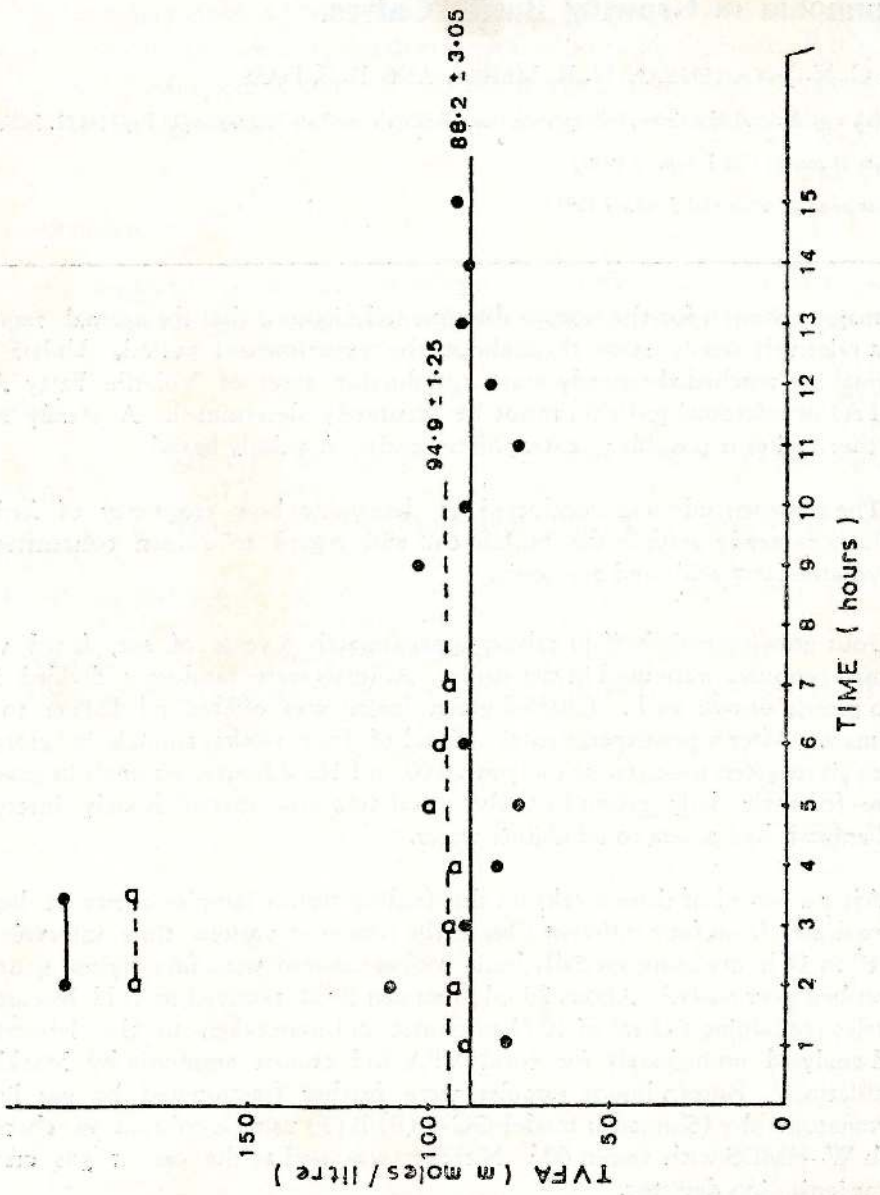


Figure 1 Total Volatile Fatty Acid (TVFA) concentration as affected by frequency of feeding in buffalo calves.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

alfalfa hay diet. Twelve times feeding at two hourly intervals in the present experiment appears to provide a more constant (94·95 ± 1·25 vs 114·8 ± 4) rumen VFA concentration compared to eight times feeding at three hourly intervals, reported earlier.[3]

The concentration and molar percentage of VFA and the concentration of rumen ammonia for the two treatments are shown in Table 1. The frequency of feeding had no influence on the concentration of total VFA although the tendency was for the animals in the twelve times feeding group to show constantly higher values. There was no significant correlation ($P<0.05$) between rumen ammonia concentration and total VFA for both feeding regimes.

Table 1. The concentration and molar percentage of Volatile Fatty Acid (VFA) and rumen ammonia.

Treatment	Rumen ammonia (mg/100ml rumen fluid)	Total VFA concentration (m moles/ litre rumen fluid)	Molar % of total VFA			Concentration of VFA		
			Acetic acid	Propionic acid	Butyric acid	Acetic acid	Propionic acid	Butyric acid
Two times feeding at 08·00 and 16·00 hours (group 1)	13·46±0·58	88·3±3·05	62·6	23·6	13·6	55·2	20·8	12·0
Twelve times feeding at two hourly intervals (group 2)	14·14±1·17	94·95±1·25	62·8	21·6	15·1	59·6	20·5	14·3

The frequency of feeding had no influence on the molar percentage of volatile fatty acids in agreement with the findings of Putnam *et al*[5] although Know and Ward[3] reported a constantly higher acetic acid percentage on the two times feeding compared to eight times feeding.

As two hourly feeding helps to maintain a constant concentration of VFA throughout the period such a feeding regime appears to be satisfactory when measuring production rates of microbial synthesis by isotope dilution techniques

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

The authors are grateful to Dr. U. B. Singh, Head Animal Nutrition Division for his interest and encouragement shown during this study. We also acknowledge the financial support received under the IAEA Fellowship programme to one of us.

References

1. CHATURVEDI, M. L., SINGH, U. B. & RANJHAN, S. K. (1973). *Indian J. Anim, Prod,* 4 (4) 194.
2. ERWIN, E. S., CARCO, G. J. & EMERY, E. M. (1961). *J. Dairy Sci.* 44 : 1768.
3. KNOW, K. L. & WARD, G. M. (1961). *J. Dairy Sci.* 44 : 1550.
4. MARKHAM, (1942). *Biochem. J.* 36 : 790
5. PUTNAM, P. A , GUTIERREZ, J. & DAVIS, R. E. (1961). *J. Dairy Sci.* 44 : 1364.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1) :

SHORT COMMUNICATION

Anuran Responses To Signal Attenuation In Environments Of High Noise

F. R. SENANAYAKE

Department of Wildlife and Fisheries Biology, University of California, Davis, California 95616, U. S. A.

(Date of receipt: 10 June 1980)
(Date of acceptance: 26 January 1981)

Anuran vocalizations, especially breeding calls in the sense of Bogert,[1] have been demonstrated to function as effective isolating mechanisms in some groups.[12] It has been shown that isolation is not the only role of this call; it may serve various other functions such as advertising the presence of a potential mate, synchronizing mating activity,[13] serving as a sex attractant,[7] or increasing female receptivity.[10] For each role the call must transmit information; indeed, the emitter and the receiver of the call represents a co-evolved unit.[3,6]

Calls are species specific and often used as taxonomic characters.[4] Studies on call structure seem to suggest that the calls may demonstrate common, discernable patterns within closely related groups of Anurans. The relationship demonstrated by such features are only consistent at the generic level, but lose validation of call characteristics with other phenomena such as habitat differences and types of mating call chorus, suggesting that these factors must exert a strong selective presence on call evolution.[9] These conditions require the shape of the call to reflect a compromise among the multiple pressures being exerted upon it.[7]

We can then define 'competitive' environments, as those in which signal intelligibility is lost, either through auditory saturation or sound attenuation. Thus we could expect species in a slightly 'competitive' environment to have call elements that demonstrate its phylogeny much clearer than species in a highly 'competitive' environment.

Animals suggest three ways in which acoustic interference leading to loss of signal information, may be reduced: frequency separation, spatial separation and temporal separation.[12] These are interspecific responses that usually occur in an environment in which two or more species call at the same spatio-temporal point. However, the physical characteristics of the environment may also influence the character of the call. Sound attenuation in amphibian voices is a function not only of call frequency but also of the physical character of the environment.[8] Other factors, such as wave reflections[2] and habitat complexity[11] have been considered. Yet another feature of the sound environment that should effect the evolution of the call structure, is the base level of background noise, termed basal noise bands, which would be a characteristic of any natural environment.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Basal noise bands would constitute abiotic noise that is sufficiently common in a given environment to provide a selective pressure on the information transmission system. Its occurence would suggest that all information transmission systems are operating at some energetically higher or more complex degree than in a free field. The increase would be rather insignificant when the basal noise level is low and the effect minimal, relative to their selective forces. When it represents a major selective force, evolutionary response to it should be demonstratable.

Some environments of high ambient noise; i.e., waterfalls and torrents, may be good breeding habitats for amphibians. Their utilization requires a solution to the problems of sound attenuation. To utilize the immediate area of such an environment, a breeding unit utilizing auditory communication could develop three basic strategies to overcome attenuation and signal loss.

The first strategy would be to develop a call of higher intensity or greater sound pressure level than background noise. This strategy may be used when the maximum background noise is lower than the biotic potential of the organism. It is also useful in environments that demonstrate a temporal variance, interspersing periods of low noise with some regularity. As every environment has some degree of random noise associated with it, all Anuran calls should incorporate response to this basal noise level. The contrasting calls of the 'quiet' frogs of the forest floor with the 'loud' calls of frogs of the African Savannah[15] may demonstrate this principle.

A second possible strategy would be to call at a higher or lower frequency than that of the ambient noise. This would be useful if the noise has a higher degree of predictability in terms of the frequency and temporal pattern. The utilization of this strategy may be demonstrated in aggregations, the species of which utilise unoccupied frequency bands to minimize call overlap with other sympatric species calling at the same time.[16]

The third strategy would be to send the call on band widths within the noise environment, but to codify the call elements. This may be accomplished by frequency modulation and amplitude modulation or both, of which there are multiple examples. Also available are the temporal units; namely call pulse rates and rate pattern which would enhance recognition without affecting attenuation.

The sound environment of the breeding habitats of the non-arboral Anurans of Sri Lanka was analyzed. (Table 1). A qualitative measure of the degree of expression of the random noise was obtained by ranking signal environments as a function of the percentage of noise occupying the sonogram trace. For the purpose of the study, areas of high ambient noise were those that demonstrated noise bands in the region of 0-5 KHz, which essentially overlaps the normally utilized band of Anurans.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table 1 – The sound environment of habitats utilized by breeding frogs in Sri Lanka

Noise Level	High	Medium	Low
Habitat Type	Wet Cliff Rapid stream Brook	Slow – Stream	River Permanent Pond Temporary Pond
Noise Expressed As Percentage Of Sonogram Trace	Over 50%	25% – 50%	0 –25%
Anurans Record	*Nannophrys ceylonensis* *Bufo kelaartii* *Rana (Hylarana) temporalis*	*Rana corrugata* *Rana greeni* * *Rana c. cyanophlictis* *Rana (Hylarana) gracilis* *	*Rana tigrina crassa* *Rana (Hylarana) aurantiaca* *Rana (Tomopterna) breviceps* *Rana hexadactyla* *Rana l. limnocharis* *Ramanella palmata* *Ramanella variegata* *Microhyla zeylanica* *Microhyla rubra* *Microhyla ornata* *Uperodon systoma* *Caloula pulchra taprobanica* *Bufo melanostictus* *Bufo fergusonii* *Bufo atukoralaei*

All Anurans calling from areas of high ambient noise utilise note modulation, suggesting some degree of correlation between the occurrence of modulation and the sound environment (Figures 1-3).None of the Anurans recorded from areas of low noise exhibit this characteristic. Note modulation itself is not an uncommon character among Anurans. It has been demonstrated in most Leptodactylids.[5] Only *Bufo quericicus* shows it in the Bufonids[14] and in the Ranids it has been demonstrated only in *Rana nigrovittata*.[9] *Bufo quericicus* does not breed in an area of high ambient noise, nor do many of the Leptodactylids

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

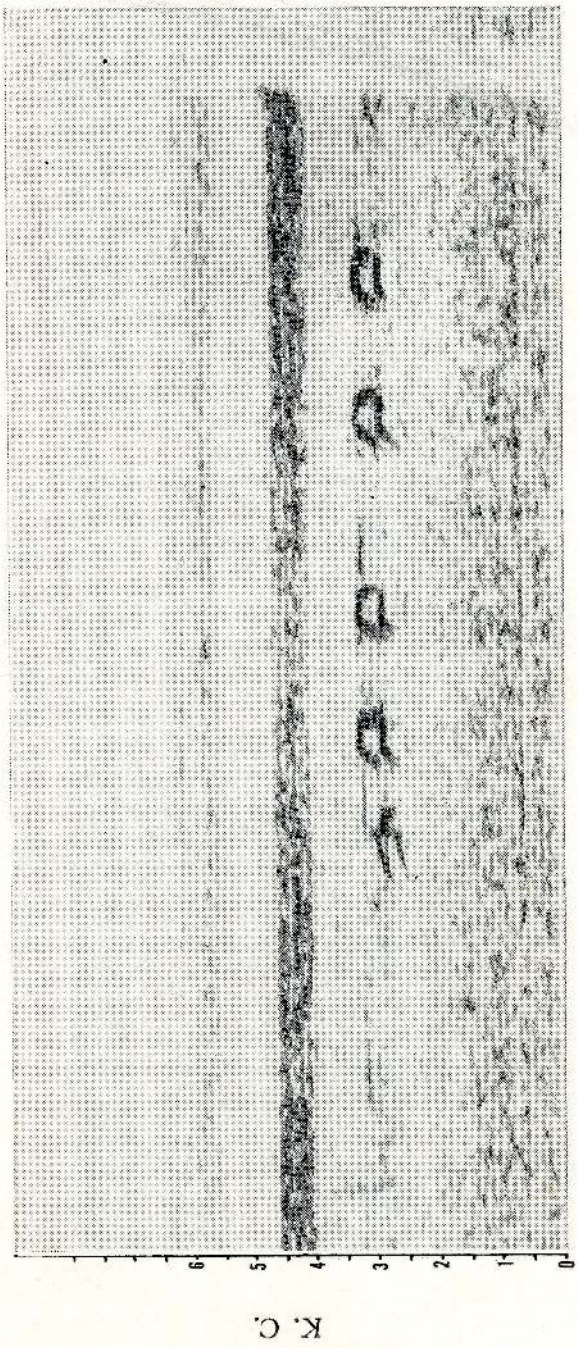


Figure 1 – Call of *Bufo kelaarti*, Kanneliya Forest 5. 16. 74, 7.00 pm; Air Temperature 26°C

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

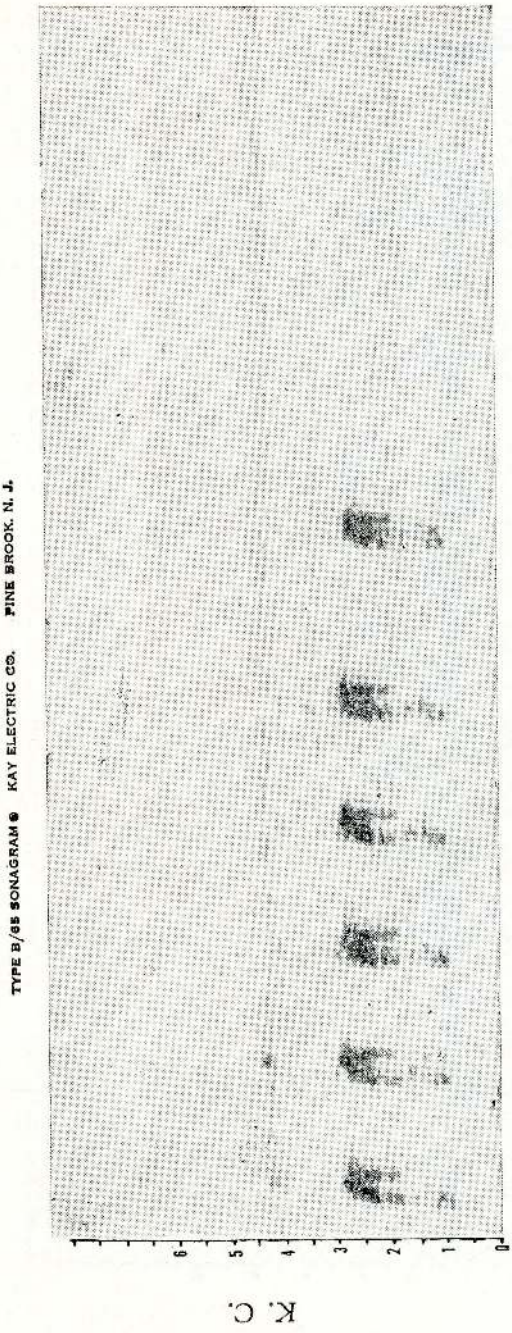


Figure 2 – Call of *Nannophyrs ceylonensis* Labugama 5. 28. 75, 7.30 pm; Air. Temperature 27°C

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

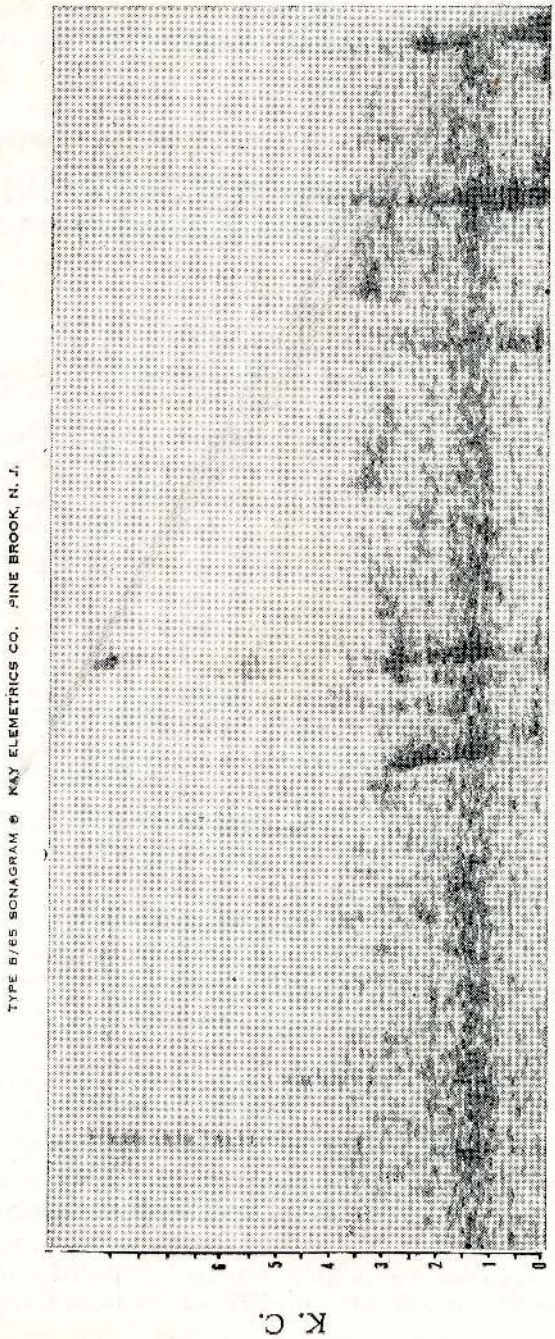
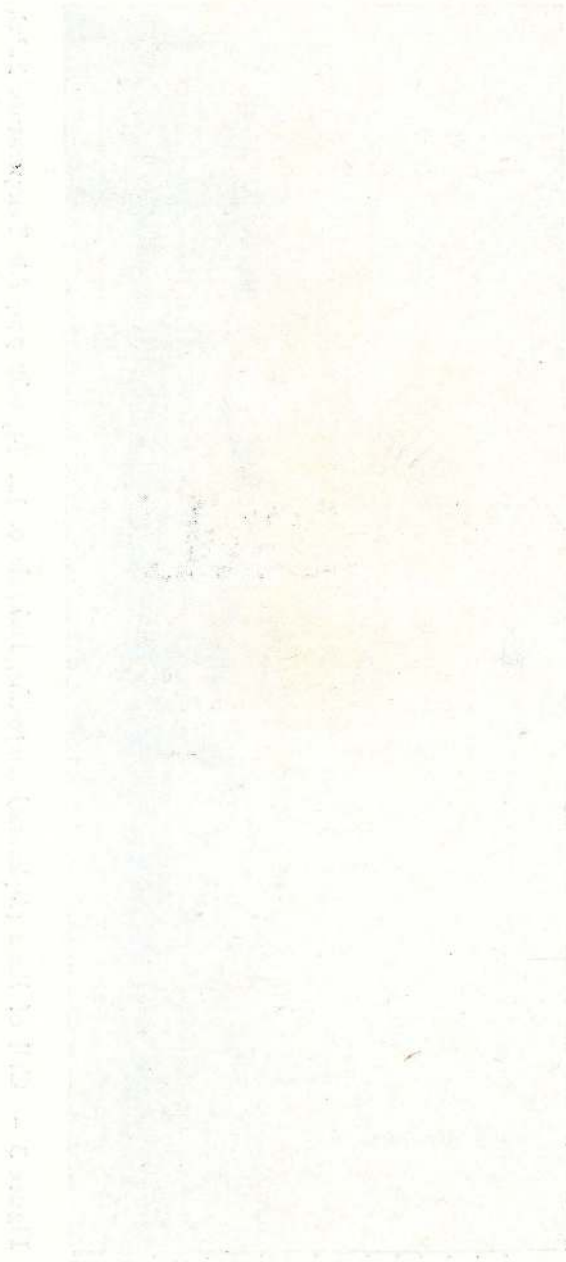


Figure 3 – Call of *Rana (Hylarana) temporalis*, Hakgala 6. 12. 74, 6.30 pm; Air Temperature 22°C.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media



Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

described by Fouquette.[5] Thus, note modulation may have other causes than high ambient noise, although its high frequency in such environments indicates its adaptive significance in overcoming high levels of background noise.

Acknowledgements

I wish to thank Prof. Carl Gans of the University of Michigan for his helpful comments and criticisms during the preparation of this paper.

References

1. BOGERT. C. M. (1960) *The influence of sound on the behavior of amphibians and reptiles.. In Animal sounds and communication*, W. E. Lanyon and W. N. Tavolga eds·*Amer. Inst Biol. Sci.* Publ. 7 Washington D. C. p. 137—320.

2. BUSNEL. R. (1977) *Acoustic Communication* In *How animals communicate*. T. A. Sebok ed Indiana Univ. Press·Bloomington and London p 233—251.

3. CAPRIANCA, R·R, (1965) *The evoked vocal response of the Bullfrog resmonograph* 33 Mass. Univ. press. Cambr. Mass. p. 1—110·

4. DUELLMAN, W·E *(1968) The taxonomic status of some American Hylid frogs. Herpetologica* **21** : 124—207.

5. FOUQUETTE, M. J. (1960) *Call structure in frogs of the family Leptodactylidae. Texas J. Sci* **12** : 201—215.

6. FRITKOFF, L·S·& N. H·GOLDSTEIN jr· *(1963) Responses to acoustic stimuli from single units in the eighth nerve of the Bullfrog. Journ. Acoust. Soc. Amer.* **35** : 1219—1228·

7. GANS, C. *(1974) Sound production in the Salientia, Mechanism and evolution of the emitters Amer. Zoll* **13***(4)* :1179—1194·

8. GERHARDT, H·C. *(1975) Sound Pressure Levels and Radiation Patterns of the Vocalizations of some North American Frogs and Toads. Journ. Comp. Physiol. A. Sens. Neural Behav.* **102***(1)*: 1—12·

9. HEYER,W·R·(1971) *Mating calls of some frogs from Thailand·Fieldiana. Zool·***58** *(6)*:61—82·

10. HUESSER, L· (1968) *Die Lebensweise Der Erderote bufo bufo* (L); *Laichzet, umstimmung ovulation, Veirteljahres, Naturforsch, Ges Zurich* **113** *(3)*:257—289

11. KONISHI, M· (1970) *Evolution of design features in the coding of species specificity. Amer. Zool* **10***(1)* :67—72·

12. LITTLEJOHN, M· J· & MARTIN, A·A· (1969) *Acoustic interaction between two species of Leptodactylid frogs. Anim. Behav.* 17:785—791·

13. MAYR E·, (1963)*Animal Speciation and Evolution. Cambridge P·i—xv*, 1—797.

14. MARTIN, W·F. (1972) *Evolution of Vocalization in the genus Bufo In evolution in the genus Bufo.* W·F·Bair (ed) Univ. of Texas Press, Austin Texas p 459·

15. SCHILZ, A· (1973) *Evolution of Anuran mating calls Ecological Aspects, In Evolutionary biology of the Anurans*, J·C·Vial (ed) Proc 13 th annual meeting of the Study of Amphibians and Reptiles·Kansas City Mo·1970;Univ·Missouri Press, Columbia Mo·p 322—327·

16. STRANGHN, I. R· (1973) *Evolution of Anuran mating calls, Bioacoustical aspects· In Evolutionary biology of the Anurans*, J· C· Vial (ed) Proc. 15th annual meeting of the Soc· for the study of Amphibians and Reptiles· Kansas City Mo· 1970; Univ· Missouri Press, Colombia Mo· p. 311 – 319·

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1) :

SHORT COMMUNICATION

The Aloin Content of Local Aloe Species

E. R. JANSZ, VAJIRA SILVA AND DAMAYANTHI RATNAYAKE

Natural Products Section, Ceylon Institute of Scientific and Industrial Research (CISIR), P. O. Box 787, Colombo 7, Sri Lanka.

(Date of receipt : 23 July 1980)

(Date of acceptance : 19 March 1981)

Aloin obtained from *Aloe* species is a mixture of anthrone glucosides and is commonly used as a laxative.[4] The genus *Aloe* grows profusely, unattended, in the Northern Province of Sri Lanka, notably in the Jaffna Peninsula and the island of Mannar. There are apparent chemical differences in the juice of samples of *Aloe* collected from those two locations although both belong to *Aloe vera* Linn. var *littoralis* Koen[2] (synonym *A. barbadensis L.*). and in this communication they will be termed Jaffna and Mannar *Aloe*.

A previous communication[2] described, (i) the identification of the plant, (ii) the solution to the Browning reactions of Jaffna *Aloe* samples, (iii) a modified method of preparation of aloin from *Aloe* juice and (iv) the isolation of an aloin (M. P. 136° - 138°C) from Jaffna *Aloe* juice. In the above investigation, assays were done by the Fairburn method.[1]

In this communication, the screening of the Jaffna and Mannar *Aloe* for aloin content using the tlc-spectrophotometric method of McCarthy[3] (which affords the accurate and rapid assay of small quantities of material) is reported along with the use of this test to monitor process losses in the isolation of aloin.

In the screening studies, juice from the 5th, 6th and 7th youngest leaves of each plant (where yield of juice is a maximum) is dissolved in methanol prior to use in the tlc technique. The solvent used was $CHCl_3$: Abs. C_2H_5OH (3:1) and aloin was estimated at 360 nm spectrophotometrically.

Assay of aloin content of the juice of individual plants showed that although there was some variation between plants of the same location, the fresh juice of the Mannar *Aloe* samples had a significantly higher aloin content than those of the Jaffna *Aloe*. (Table I)

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Table I – Main differences between Jaffna and Mannar Aloe

Place	Aloin content %		Resin content %
	Fresh juice	Dried product ("aloes")	
Mannar	16·5	57	15
Jaffna	9·0	30	30

The above refers to bulk samples.

In the Jaffna locations (Kayts, Mandaithivu) aloin content of individual plants was in the range of 8% to 12% while in the Mannar plants (which were studied in more detail) the variation between plants sampled at random were as in Table 2. The aloin content of plants in Vidatheltivu and Poonakay (location situated on the coast between Jaffna and Mannar) was between 11% and 12%.

Table 2 – Variation in Aloin content of Mannar Aloe

Percentage Aloin	< 12·5	12·5 — 15	15 — 17·5	17·5 — 20	> 20
Number of plants	3	6	13	5	2

The Jaffna 'aloes' resemble commercial 'aloes' not only in its aloin content but also in resin content (water insoluble material). The Mannar 'aloes' are outstanding in its mean aloin content of 57% (dry basis) being at least 50% more than the highest reported in the literature.[3,4,5,6] Further its resin content is abnormally low. In fact the tlc pattern (sprayed with fast blue and KOH) of fresh aloe juice (Mannar) resembles semi-purified aloin rather than fresh aloe juice due to its low concentration of resins and resin precursors.

Seasonal variations were not studied in detail but it was noticed that season affected output of juice rather than aloin content of bulk aloe samples. The same is true for maturity of plant where there was no significant difference in aloin content but the quantity of aloe juice increased with maturity. An interesting finding was that plants growing in the shade had the highest aloin content.

Quantitative tlc analysis of the products at different stages of isolation of aloin from "aloes" showed that of the aloin available, nearly three fourths could be isolated; the losses being mainly due to destruction during processing and unrecoverable in supernatants (15% to 20%) and co-precipitated in resin (8 % to 10%).

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Both Jaffna and Mannar "aloes" gave a single, identical and unresolvable aloin spot on tlc analysis. Isolation of this aloin resulted in lemon-yellow crystals having an $[\alpha]_D^{30}$ value of $-1 \cdot 9$ (methanol) thus differing from barbaloin and its optical isomer at C[1] (isobarbaloin) which have reported values of $+21$ and -19 respectively.[4] The possibility of this being a mixture of the two compounds appears unlikely as solvents capable of separating the two on tlc fail to do so. The melting point of the aloin isolated was 136° C to 138°C[2] which differs from that reported in the literature for barbaloin (148° C to 148·5° C).[4]

Aerial oxidation of the aloin to the corresponding anthraquinone showed that the latter on isolation had an I. R. spectrum (γ max, 3400 and 1620) consistent with a 1 : 8 dihydroxyanthraquinone (as is aloe-emodin, the anthriquinone derived from barbaloin and isobarbaloin) thus showing that this aloin is structurally closely related to those reported previously. It appears possible that any structural difference may lie in the sugar moiety. Further spectroscopic investigations are in progress.

These studies have shown that the Mannar *Aloe* are a better source of aloin than Jaffna *Aloe* for the following reasons:

(a) More raw material is available.
(b) Yield of juice (>lg/leaf) is nearly double that of Jaffna
(c) Aloin content is higher.
(d) Lower resin content (would facilitate extraction of aloin)
(e) Browning reaction is less prevalent in Mannar "aloes" (lighter, more marketable product).

It is not clear if the differences observed are due to genetic or environmental factors or a combination of both.

Acknowledgements

The authors thank Mr. S. R Krishnarajah for hosting us on our field trip to Jaffna; Dr. S. Sentheshanmuganathan of the Medical Research Institute (MRI) for the use of his spectrophotometer, Mr. H. M. Dharmadasa, Mr. S. Balachandran and Miss S. Wijeratne for technical assistance and Mrs. Devika Tennekoon and Miss Dinapali de Zoysa for secretarial assistance.

References

1. FAIRBURN, J. W. & SIMIC, S. (1963) *J. Pharm. Pharmac.* 15 : 325
2. JANSZ, E. R., JEYARAJAH, E. J. ET AL. (1980) *Proc. I. Chem. C.* (Sri Lanka). 9th Annual Session, part 1, p. 2.
3. McCARTHY, T. J. & MAPP, R. K. (1970) *Planta medica* 18: 36 43.
4. THOMSON, R. H. (1971). *In "Naturally occurring quinones"* p. 399–400 2nd edition Acad. Press – London.
5. VERMA, N. (1971) *I.O and S.J.* Sept. 1971, p. 63 – 65.
6. Wealth of India, *Raw Materials*, p. 60—62.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1):

ලිපිවල සාරාංශ - සිංහල පරිවර්තන

---

(1) ඇතැම් මුදු ඇල්ගී වර්ගවල ඇල්ජිනික් අම්ල සහ ඒගාර් අන්තර්ගතය පිළිබඳ ප්‍රාථමික අධ්‍යයන.

ඉන්ද්‍රාණි ආරුමුගම්, ඒ. සිවසාළන් සහ කේ. තෙයිවේන්දිරරාජා

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1):

දුඹුරු ඇල්ගී වලින් ඇල්ජිනික් අම්ල නිස්සාරණය කිරීමේ විවිධ ක්‍රම අත්හද බලන ලදුව $Na_2 Co_3$ සහ ඉන් අනතුරුව විරංජන කුඩුවලින් පිළියම් කිරීමෙන් තත්වයෙන් වඩා උසස් ඇල්ජිනික් අම්ල ලැබෙන බැව් හෙළිවිය. අධ්‍යයනයට භාජනය කරන ලද දුඹුරු ඇල්ගී වර්ග අතුරින්, වැඩිම ඇල්ජිනික් අම්ල ප්‍රමාණයන් අන්තර්ගත වූයේ *Cystoseira triquetra* (L) J. Agardh සහ *conoides* (Kuetzing) යන වර්ග වලය. මෙම ඇල්ගී වර්ග වල අන්තර්ගත ඇල්ජිනික් ප්‍රමාණය සෘතුවෙන් සෘතුවට සැළකිය යුතු අන්දමින් වෙනස් වන බව පරීක්ෂණ වලින් තවදුරටත් පෙනී ගියේය. වැඩිම ප්‍රමාණයක් පැවතියේ මැයි හූලි දක්වා මාස වලය. එකිනෙකට වෙනස් රතු ඇල්ගී විශේෂයන් 6 කින් ඒගාර් නිස්සාරණය කරන ලදුව, ඒගාර් නිස්සාරණය කිරීම සඳහා *Gelidium* (Lamouroux), Hypnea (Lamouroux සහ Gracilaria (Grev ille) යන විශේෂයන් යොදගත හැකි බැව් හෙළි විය. මේවායේ අන්තර්ගත ඒගාර් ප්‍රමාණය 16·2% ත් 50% අතර විය. මෙම ඇල්ගී වර්ග වල ඇතුලත් ඒගාර් ප්‍රමාණයේ සෘතුවෙන් සෘතුවට සැළකිය යුතු වෙනසක් තිබිණ. ජනවාරි මාසයේදී ඒවායේ අඩංගුවන ඒගාර් අන්තර්ගතය අධික බැව් පෙනී ගියේය.

---

(2) ශ්‍රී ලංකාවේ මුහුදු ඇල්ගී වර්ගවල කාබෝහයිඩ්‍රේට් සංඝටක.

1. කොටස. රතු ඇල්ගී වර්ග අටකින් ලබාගත් ඇල්ගී කලිල වල භෞත-රසායනික ගුණාංග කිපයක්.

ඒ. පී. දන්නනාරායන, එන්. සාවිත්‍රි කුමාර්, එම්. යූ. එස්. සුල්තාන් බාවා සහ එස්. බාලසුබ්‍රමනියම්.

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1):

රතු ඇල්ගී විශේෂයන් අටකින් ඇල්ගී කලිල නිස්සාරණය කර, ලැබුණු ප්‍රමාණය ඇස්ත-මේන්තු කරන ලදී. වාණිජමය වශයෙන් ප්‍රයෝජනයට ගැනීම සඳහා සුදුසුතම වර්ග ලෙස පෙනී ගියේ *Gracilaria edulis* (*lichenoides*) සහ G. *salicornia* යන විශේෂයන්ය. මේවායේ ජෙල ප්‍රබලතා ගුණයන්ද, ඇල්ගී කලිල වල රසායනික සංඝටක සහ එම ගුණයන් අතර ඇති සම්බන්ධයද, වාණිජමය ඒගාර් සාම්පලයක සමාන ගුණාංගයන් ඇසුරින් සාකච්ඡා කර ඇත.

පූර්ණ සංඛ්‍යාමය මධ්‍යනායක අභිසාරිතා සාධක

සී. යෝගචන්ද්‍රන්

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1);

f,g ශ්‍රිතවල සුදුසු සීමා කිරීම් යටතේ සහ rkEIo, $r \geqslant k \geqslant O, p, q \in R$ සහ $p+q$ $\{-1, -2, \ldots\ldots\ldots -r\}$ යන උපකල්පනය යටතේ, $\lim_{k \to f(x)} f(x)$ හි K වෙනි පුනරාවර්තක පූර්ණ සංඛ්‍යාවක් නිරූපණය කරයි නම්;

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

$I_K kf(x) = \sum_{m=o}^{R-I} c_m x^{k-l-m} + O\left(x^{p+k+r}\right)$ වනවිට එක්තරා තාත්වික $C_m(^{mk=o.I. \ldots r-l})$

සඳහා

$I_r+(x)\, gc\, x) = \sum_{m=o}^{r-i} c_m x^{r.l-m} + O\left(x^{p+q+r}\right)$ වීමට අනිවාර්ය හා ප්‍රමාණවත් අවශ්‍යතාවයන් මෙම ශීර්ෂයෙන් ඔප්පු කරනු ලබන ප්‍රධාන ප්‍රමේයයෙන් දෙනු ලබයි.

ඇෆ්ලටොක්සින් bl වලින් පිළියම් කළ *Rhacophorus leucomystax maculatus* (Gray) වර්ගයේ ඉස්ගෙඩියන්ගේ අක්මාව පිළිබඳ ඉලෙක්ට්‍රෝන අන්වීක්ෂීය පරීක්ෂණයක්.

ඒ. ඩී. පී. ජයතිලක සහ එස්. කිරුපනන්දන්.

J. Natn. Sci, Coun. Sri Lanka 1981 **9** *(1):*

ඉස්ගෙඩියන් පියවි ඇසට දිස්වන බැවින්, කලල ද්‍රව්‍ය නිරීක්ෂණය සඳහා ඔවුන් තෝරා ගැනිණ. *Rhacophorus leucomystax maculatus* ඉස්ගෙඩියන්ට ඇෆ්ලටොක්සින් bl වල LD50 යොදා පිළියම් කරන ලදී. පිළියම් කිරීමෙන් පසු දෙවන 3 වන සහ 4 වන දිනවල ඉස්ගෙඩියන් මරණාසන්න වූහ. පිළියමින් නොමැරී ඉතිරිවූවන් අළුත් ආසුන ජලය කුලට මාරු කරන ලදී. පාළන ඉස්ගෙඩියන්ගේ සහ 2වන, 3වන සහ 4වන දිනයන්හි මරණාසන්න වූවන්ගේ අක්මා, සූක්ෂ්ම විච්ඡේදනයට භාජනය කර, පිරිසැකසුම්කර ඉලෙක්ට්‍රෝන අන්වීක්ෂයක් මඟින් නිරීක්ෂණය කරන ලදී. පිළියම් කිරීමෙන් පසු නොමැරී ඉතිරිවූණු ඉස්ගෙඩියන්ගේ අක්මාද සති දෙකකට පසු පිරිසැකසුම් කරන ලදී. පිළියම් කළ ඉස්ගෙඩියන්ගේ හෙපැටොසයිට වල න්‍යෂ්ඨිකා කුළුවැස්මක් විය. සෛලප්ලාස්මයේ මේදමය ආස්‍රාවනයක් විය. ඉන්ද්‍රිකාවල ව්‍යුහමය වෙනස්කම් දක්නට ලැබුණ අතර ස්ඵටික සහිත බවද, ග්ලයිකොජන් වලින් මුළු මනින්ම තොර බවද පෙණිනි. පාළන ඉස්ගෙඩියන්ගේද, පිළියමින් පසු නොමැරී ඉතිරි වූ ඉස්ගෙඩියන්ගේද හෙපැටොසයිටවල ග්ලයිකොජන් පැවතිණ. පිළියමට භාජනය කළ ඉස්ගෙඩියන්ගේ හෙපැටොසයිටවල ඇතිවූ ව්‍යුහමය වෙනස්කම් වලට හේතුව, සෛල කෙරෙහි ඇෆ්ලටොක්සින් වලවූ විෂ ගතිය විය හැක.

---

(5) භූතල ජල සම්පාදන ක්‍රම වලට ඇතිවන භෝග ප්‍රතිචාරය

එන්. සේනානායක

J. Natn. Sci. Coun. Sri Lanka 1981 **9** *(1):*

1973–1974 වර්ෂයේදී, භෝග ප්‍රතිචාරයට අදාලව, වාරිමාර්ග ක්‍රම පිළිබඳ සන්සන්දනාත්මක අධ්‍යයනයක් කරන ලදී. ඇගැයීමට ලක් කරන ලද වාරිමාර්ග ක්‍රම දෙක අතුරින් සම්මත ඇලි ක්‍රම හා සසඳා බලන කල අක්වක් ඇලි ක්‍රමයට කැපී පෙනෙන තරම් වැඩි භෝග ප්‍රතිචාරයක් දක්නට ලැබිණ. මෙම වාරි මාර්ග ක්‍රමය අනුව අධික නෙතමන ප්‍රතිචාර භෝගයන්ගෙන් ලැබෙන අස්වැන්න කැපී පෙනෙන සුළු විය. අක්වක් ඇලි සම්බන්ධයෙන් දක්නට ලැබුණු තවත් ලක්ෂණයක් නම් සම්මත ඇලි හා සසඳා බලන කල අක්වක් ඇලි වල විවිධ කොටස් වල අස්වැන්න වඩා ඒකාකාරී වීමයි.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

ප්‍රධාන දළ ආහාරය වශයෙන් ඇල්කලි යොදා පිළියම් කළ පිදුරු දෙනු ලබන බැටළුවන් සඳහා තේ රොඩුවල සහ යූරියාවල ඇති නයිට්‍රජන් උපයෝගීකරණය.

එම්. සී. එන්. ජයසූරිය

J. Natn. Sci. Coun. Sri Lanka 1981 **9** (1) :

යෝඩියම් හයිඩ්‍රොක්සයිඩ් වලින් පිළියම් කළ පිදුරු වලට ප්‍රෝටීන් එක් කිරීමේ මාධ්‍යයක් වශයෙන් තේ රොඩු (STL) වල ඇති ප්‍රයෝජනවත් බව පරීක්ෂා කිරීම සඳහා දෙමුහුන් බැටළුවන් පෝෂණය කිරීමේ අත්හදා බැලීමක් පිළිබඳව වාර්තාවේ. අඹරන ලද බඩ ඉරිඟු සමඟ යූරියා (සාන්ද්‍රන ග්‍රෑම් 100 ට ග්‍රෑම් 14 ක් හෝ 18 ක්) හෝ තේ රොඩු (සාන්ද්‍රන ග්‍රෑම් 100 ට ග්‍රෑම් 18 ක්) මිශ්‍ර කිරීමෙන් පිළියෙල කළ සාන්ද්‍රන කොටයක් තුනක් සන්සන්දනය කරන ලදී. ආහාර රුචිය ඇති කිරීම සඳහා දළ ආහාර වශයෙන්, පිළියම් කළ පිදුරු (බ/බ 4%) පමණක් දෙන ලදී. තේ රොඩු අන්තර්ගත වූ ආහාරයන්හි දළ ප්‍රෝටීන් ප්‍රමාණය 6% කට සීමා වූව ද, තේ රොඩු මිශ්‍ර ආහාරය ගත් සතුන්ගේ වැඩීම, අප්‍රෝටීන් නයිට්‍රජන් යූරියා ආහාරය ලැබුවන්ගේ තරමටම සතුටුදායක විය. මෙම නිරීක්ෂණය අනුව තේ රොඩු (STL) ප්‍රෝටීන් වල ජීරනය නොවන සුවභාවයක් තිබිය හැකි බැව් පෙන්නුම් කෙරෙයි. එය යෝඩියම් හයිඩ්‍රොක්සයිඩ් වලින් පිළියම් කළ පිදුරු මූලික ආහාරයන් සඳහා අනුපූරක ප්‍රෝටීන් සැපයීමේ අගනා මාධ්‍යයක් ද වෙයි.

දත් දිරා යාමේදි බෙටික නිර්ද්‍යාස ග්ලෝබියුලින සහ ලැක්ටොෆෙරින්.

එස්. දිසානායක එල්. පී. සමරනායක සහ ආර්. එම්. බෙනට්

J. Natn. Sci. Coun. Sri Lanka 1981 **9** (1) :

අධික ලෙස සහ මද වශයෙන් දත් දිරා යාමට භාජන වූවන්ගේ මස්තු $I_g G$, බෙටික $I^g A$, සහ බෙටික ලැක්ටොෆෙරින් මට්ටම් නිර්ණය කරන ලදී. අධික ලෙස දත් දිරා යාමට පාත්‍ර වූවන්ගේ නිර්ද්‍යාස ග්ලොබියුලින සහ ලැක්ටොෆෙරින මට්ටම් ඉහල වන බැව් හෙලි විය.

ශ්‍රී ලංකාවේ, ස්පර්ශයෙන් බෝවන ස්කේබීස් රෝගය සහ දරුණු ගුවිශිකාවාක්කප්‍රදාහය ; පූර්ව සහ පශ්චාත් අධ්‍යයනයක්.

එස්. එන්. අර්සකුලරත්න, එන්. ශර්මනපවන්, සී. නවරත්නම් සහ ඩී. ඒ. ගුණවර්ධන

J. Natn. Sci. Coun. Sri Lanka 1981 **9** (1) :

ස්පර්ශයෙන් බෝවෙන චර්ම රිෂ්ට සහිත දරුණු ගුවිශිකා වෘක්ක ප්‍රදාහය හේතු කොටගෙන ආරෝග්‍යශාලා ගත කරන ලද රෝගීන් පිළිබඳ පරීක්ෂණයකදී, මෙබඳු රෝගීන් අතර වඩාත්ම ප්‍රචලිත (79%) පූර්ව ආසාදනය ස්කේබීස් බව පෙනී ගොස් ඇත. ස්පර්ශයෙන් බෝවන ස්කේබීස් රෝගය බොහෝ විට දරුණු ගුවිශිකාවාක්ක ප්‍රදාහයෙහි පූර්ව ආසාදනයක් වන බවට ට්‍රිනිඩැඩ් සහ දකුණු අප්‍රිකාවෙන් ලැබී ඇති වාර්තා මෙයින් තහවුරු වෙයි. ස්පර්ශයෙන් බෝවෙන ස්කේබීස් රෝගය වැළඳුන රෝගීන් පිළිබඳව කරන ලද පූර්ව අධ්‍යයනයක දී, මුත්‍ර පිළිබඳ අසාමාන්‍යතා ඇතිවූයේ ඔවුනතුරින් 8% කට පමණක් බැව් හෙලි විය. මෙම වෙනසට හේතු විය හැකි කරුණු මෙහි සාකච්ඡා කෙරෙයි. මෙම චර්ම රිෂ්ට වලින් වෙන් කර ගන්නා ලද රක්තානුජ්වලීක ස්ට්‍රෙප්ටොකොකුසයින්ගේ මස්තු වර්ග රටාව වෙනත් රටවලින් වාර්තාවී ඇති ඒවාට සමානය. දරුණු ගුවිශිකාවාක්කප්‍රදාහය සහිත රෝගීන්ගෙන් 40% දෙනෙකුගේ චර්ම රිෂ්ට වලින් *Staph-aureus* පමණක් ලැබිණ. එහෙත් මේවායේ දක්නට ලැබුණු පුළුල් ලෙස වෙනස් වූ ගුණාංග වලින් විශේෂ වෘක්කප්‍රදාහයික මාදිලි ඇති බවක් නොහැගවිණ. ගුවිශිකාවාක්කප්‍රදාහයෙන් පෙළෙන හෝ නොපෙළෙන රෝගීන්ගේ චර්ම රිෂ්ට වලින් වෙන් කර ගත් *Staph-aureus* අතර වැඩි වශයෙන්ම පැවතියේ III වන භක්ෂ කාණ්ඩයයි. එහි

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

වඩාත්ම ඉස්මතුවී පෙනෙන හක්ෂ වර්ගය 54 යි. වෙන් කර ගන්නා ලද ස්ටැෆිලොකොකසයන් හක්ෂ රටා ව්‍යාප්තිය අනින්, සෞම්‍ය දේශගුණික රටවලින් වාර්තාවී ඇති රටා-වනට වෙනස් විය. ප්‍රයෝ චර්මයට අනතුරුව හට ගන්නා ගුවිශීකාවෘක්ක ප්‍රදාහය සම්බන්ධයෙන්, වෙනත් රටවලින් වාර්තාවී ඇති ආකාරයේ සැහවුණු කාල පරිච්ඡේදයක් ඇති බැවි පෙනී ගියේය. ප්‍රයෝ චර්මයෙහි ප්‍රමාණය, ගුවිශීකාවෘක්ක ප්‍රදාහය වැළඳීම හෝ එහි දරුණු භාවය කෙරෙහි බල නොපායි. ප්‍රයෝ චර්මයට පසුව වැළඳෙන වෘක්ක ප්‍රදාහයෙහි ASO අනුමාපන (ටීටර) අගය සාමාන්‍ය පුද්ගලයින්ගේ සහ දරුණු රුමැටිසිය ජ්වරයෙන් පෙළෙන්නවුන්ගේ එම අගයට අතර මැදි ප්‍රමාණයක විය. පශ්චාත් ස්ට්‍රෙප්ටොකොකස වෘක්ක ප්‍රදාහය වැළඳුන රෝගීන් විකීර්ණ වශයෙන් මෙරට බහුලව හමු වුවද එය වසංගතයක් වශයෙන් මෙහි පැතිර නොමැත.

Sitotroga cerealella (Olivier) වලට දේශීය ශාක නිස්සාරිතයන්ගේ වාෂ්ප ආසුතවලඇති විකර්ශක සහ මාදක ගුණ පිළිබඳ පරීක්ෂණාගාර විමර්ශණ.

වී. කේ. ගනේසලිංගම් සහ එස්. ආර්. කිෂ්ණරාජා.

J. Natn. Sci. Coun. Sri Lanka 1981 **9** (1):

ශ්‍රී ලංකාවේ වී වලට වැළඳෙන වැදගත් පළිබෝධයක් වන *Sitotroga cerealella* වලට එරෙහිව භාවිතා කරනු පිණිස සුදුසු දේශීය ශාක නිස්සාරිත නිර්ණය කිරීම සඳහා අධ්‍යයනයක් කරන ලදී. භාවිතා කළ ද්‍රව්‍ය කෙරෙහි සලබයින්ගේ හැසිරීම නිර්ණය කිරීම සඳහා "Y" හැඩයේ කෘමි මාපකයක් භාවිතා කරන ලදී. මෙම ද්‍රව්‍ය වල වාෂ්පයෙහි ඇති ධූලක බලපෑම පිරික්සීම සඳහා විශාල පෙට්‍රි දිසිවල අතිරේක පරීක්ෂණ පවත්වන ලදී. සැමෙකකම ප්‍රතිවලිත කිපය බැගින් ගන්නා ලදී. භාවිතා කළ ද්‍රව්‍ය අතුරින් *Vitex negundo* වල අන්තර්ගත භූමාල වාෂ්පශීලී සංඝටකයන් සතුටුදායක විකර්ෂකයක් විය හැකි බවද පැහිරි තෙල් වල වාෂ්පය *Sitotroga cerealella* වලට ධූලක වන බවද, පර්යේෂණ වලින් ලැබුණු දත්තයන් සංඛ්‍යා ලේඛණාත්මක වශයෙන් විශ්ලේෂණය කිරීමේදී හෙලිවිය.

නයිට්‍රජන් භාවිතය සහ ශ්‍රී ලංකාවේ අතුරු භෝග වගා ක්‍රමයට අදල ආර්ථික සාධක.

එච්. පී. එම්. ගුණසේන.

J. Natn. Sci. Coun. Sri Lanka 1981 **9** (1):

නයිට්‍රජන් උපයෝගීකරණය සහ දැනැමි අතුරු භෝග වගා ක්‍රම වලට අදල ආර්ථික සාධක සත්සන්දනය කිරීම සඳහා, බඩ ඉරිහු / කව්පි, බඩ ඉරිහු / සෝයා බෝංචි සහ බඩ ඉරිහු / මුං යන භෝග සංකලනයන් යොදු ගනිමින්, ශ්‍රී ලංකාවේදී ක්ෂෙත්‍ර අධ්‍යයනයන් පවත්-වන ලදී. නයිට්‍රජන් යොදන ලද්දේ, රනිල නොවන භෝග වර්ග සඳහා දේශීය වශයෙන් නිර්දේශ කර ඇති ප්‍රමාණයන්ගෙන් 0%, 50% සහ 100% යන ප්‍රමාණයනට අනුවය. සෑම ස්ථානයකදීම, භෝග වගා ක්‍රම දෙක යටතේම රනිල නොවන භෝග වර්ග වලින් නයිට්‍රජන වලට ධනාත්මක ප්‍රතිචාරයක් ඇතිවීණ. වැඩි නයිට්‍රජන් මට්ටමක් සහිතව අතුරු භෝග ක්‍රම-යට වගා කළ රනිල වර්ග වල අස්වැන්න පහත වැටින. මෙයට හේතුව රනිල නොවන භෝගයෙන් සෙවන වීම විය හැක. අතුරු භෝග වාගවෙන් මූළු නිපැයුම සහ දළ ආදායම වැඩිවිය. අතුරු භෝග වගා ක්‍රමයේ ශුද්ධ ප්‍රෝටීන උපයෝගිකරණ අගයද ඉහල විය. සංවර්ධනය වන රටවල යැපුම් ගොවිනට එබඳු ක්‍රමවල ඇති වැදගත් කම ඉන් හුවා දැක්වෙයි.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

1. கடல் அல்காக்கள் சிலவற்றிலுள்ள அல்கினிக்கமிலம், அகார் அளவுகள் பற்றிய முதனிலை ஆய்வு.

இந்திராணி ஆறுமுகம், ஏ. சிவபாலன், கே. தெய்வேந்திரராஜா

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1);

கபிலநிற அல்காக்களிலிருந்து அல்கினிக் கமிலத்தைப் பிரித்தெடுக்கப் பல் வேறு முறைகள் பரீட்சார்த்தமாகக் கையாளப்பெற்றுள்ளன. Na_2, Co_3 ஆகிய பதார்த்தங்கள் கலக்கப்பெற்று வெளிற்றுந்தூளால் பண்பூட்டப்பெற்ற பின்னர் சிறந்த ரக அல்கினிக்கமிலம் பெறமுடியுமென்பது கண்டறியப்பட்டது. ஆய்வுக்கு உட்படுத்தப்பெற்ற கபிலநிற அல்காக்களில் கிறிஸ்டோ செம்ரு திரிகுலேற்ரு (எல்) ஜே அகாந்து எனப்படும் அல்கா இனமும் துர்பினுரியா கொனெய்டேஸ் (குவேற் சின்) எனப்படும் அல்கா இனமும் மிகக்கூடிய அல்கினிக் கமிலம் கொண்ட வைகளாகக் காணப்பட்டன. மேற்கொண்டு செய்த ஆய்வுகளின் பின்னர், இந்த அல்காக்களிலுள்ள அல்கினிக் கமிலம் பருவ மாற்றங்களுக்கேற்பப் பெரிதும் வேறுபடும் இயல்பு கொண்டிருக்குமென்பதும் இந்தப் பருவ வேறுபாடுகள் மே மாதம் முதல் யூலை மாதம் வரை அதிகம் நிகழக் கூடியன வென்பதும் கண்டறியப் பட்டது. செந்நிறங் கொண்ட ஆறு அல்கா வகைகள் அகார் பதார்த்தத்தினை பிரித்தெடுப்பதற்குப் பயன்படுத்தப்பட்டன. கெலீடியம் (லமோரூ), இர்ளியா (லமோரூ), கிருசிலாரியா (கிறேவில்) ஆகிய செந்நிற அல்கா இனங்கள் அகார் பிரித்தெடுப்பதற்குப் பயன்படுத்தப்படலாமென்பதும் புலனுகியது. இந்த அல் காக்களிலுள்ள அகார் அளவு 16·2% முதல் 50% வரை இருப்பதாகத் தெரி கிறது. இந்த அல்காக்களின் அகார் அளவு பருவங்களுக்கேற்பப் பெரிதும் மாற்ற முறும் இயல்புடையது. சனவரி மாதத்தில் இந்த அல்காக்களில் அதிக அள வான அகார் இருக்குமென்பதும் கண்டறியப்பட்டது.

---

2. இலங்கையின் கடல் அல்காக்களிலுள்ள காபோவைதரேற்றுக் கூறுகள். பாகம் I எட்டு வகை செந்நிற அல்காவினங்களின் கூழ்பொருள்சார் சில பௌதிகவிரசாயன உடைமைகள்.

ஏ. பி. தந்தநாராயண, சாவித்திரி குமார், பூ. எஸ். கல்தான்பாவா, எஸ். பாலசுப்பிரமணியம்

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1);

எட்டுவகை செந்நிற அல்காக்களிலிருந்து கூழ்ப்பொருள்கள் பிரித்தெடுக் கப்பட்டு அவ்வினவுகள் மதிப்பீடு செய்யப்பட்டுள்ளன. கிருசிலேரியா ஏடி யுலிசு (இலெக்கனேயிடேசு), எனப்படும் இனமும் ஜீ. சலிக்கோர்னியா எனப் படும் இனமும் வர்த்தகமுறை உபயோகத்திற்கு உகந்த அல்காக்களாக இருக்கின்றன. செல் வலு உடைமைகள் அல்காக் கூழ்ப் பொருள்களின் இரசாயனக் கூறுகளுடன் கொண்டுள்ள தொடர்பானது வர்த்தகமுறை உபயோகத்திற்கியன்ற அகார் மாதிரி யொன்றிலுள்ள அதே இயல்புடைய உடைமைகளுடன் ஒப்பு நோக்கி ஆராயப்பட்டுள்ளன.

---

3. தொகையீட்டு இடையுறுப்பொன்றின் ஒருங்கற் காரணிகள்.

சி. யோகசந்திரன்

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1):

இக்கட்டுரையில் நிறுவப்பெற்றுள்ள தேற்றமானது f, g சார்புகளைக் கட் டுப்படுத்தும் சாதகமான தடுப்புகளின் கீழும் r, k E $\mathbb{N}_0$, $r \geqslant k \geqslant o$, p, qER உம் $p+q \notin \{-1, -2, -\cdots -r\}$ உம் ஆகிய எடுகோள்களின் கீழும் $I^{k} f(x)$ என்பது $f(x)$ இன் kஆம் மீளச் செய்த தொகையீட்டினைக் குறிப்பதான

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

$$I_k f(x) = \sum_{m=0}^{k-1} c_m x^{k-1-m} + O\left(x^{p+k+r}\right) \quad \text{ஆகவும்}$$

$$I_r f(x)g(x) = \sum_{m=0}^{r-1} c_m x^{r-1-m} + O\left(x^{p+q+r}\right) \quad \text{ஆகவும்}$$

இருக்கும்போது மெய்யான c_m (m O, 1, ... r−1) சிலவற்றிற்கு வேண்டிய போதிய நிபந்தனைகளைக் கொடுக்கின்றது.

---

4. அப்லாதொட்சின் பீளல் தொழிற்படவிடலுக்கு உட்படுத்தப் பட்ட குகோபோறஸ் லியுகோமிஸ்டாகஸ் மகுலாடுஸ் (கோறை) இனம் சார்ந்த வாற்பேய்களின் ஈரல் மீதான இலத்திரன் நுணுக்குக்காட்டி சோதனைகள்.

ஏ. டீ. பீ ஜயதிலக்கா, எஸ். கிருபானந்தன்

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1):

வெறும் கண்ணுக்குப் புலப்படும் வாற்பேய்கள் முளையப் பொருள்களைக் கவனிப்பதற்கு தேர்ந்தெடுக்கப்பெற்றன. குகோபோறஸ் லியுகோமிஸ்டாக்ஸ் மர்குலாடுஸ் என்னும் இனம்சார்ந்த வாற்பேய்கள் எல்டீ 50 அப்லாதொட்சின் பீளல் மருந்தில் தொழிற்படவிடப்பெற்றன. தொழிற்பட்விடலின் 2, 3, 4 ஆகிய நாட்களுக்குப் பின் வாற்பேய்கள் இறக்கும் தறுவாயில் இருந்தன. உயிர் பிழைத்த வாற்பேய்கள் புதிய வடித்த நீரில் இடப்பட்டன. சோதனை வாய்பேய்களினதும் 2, 3, 4 ஆகிய நாட்களில் இறக்கும் தறுவாயில் இருந்த வாற்பேய்களினதும் ஈரல்கள் நுண்ணறுக்கல் முறையால் அறுக்கப்பெற்று சீர் செய்து இலத்திரன் நுணுக்கக் காட்டி மூலம் அவதானிக்கப்பட்டன. தொழிற்படவிடலுக்கு உட்பட்டு உயிர் தப்பியவற்றின் ஈரல்களும் இரண்டுவாரங்களுக்குப் பின் சீர்செய்யப்பட்டன. தொழிற்படவிடலுக்கு உட்பட்ட வாற்பேய்களின் ஈரல் குழியங்களில் புன்கருமுகடாதல் காணப்பட்டது. குழியமுதலுருவில் கொழுப்பு ஊடுபரவி இருந்தது. புன்னங்கங்களில் கட்டமைப்பு மாற்றங்கள் ஏற்பட்டிருந்தன. பலிங்குக் கட்டிகள் காணப்பெற்றதுடன் கிளைக்கோசன் அறவே இல்லாதிருந்தது சோதனை வாற்பேய்களினதும் தொழிற்படவிடலுக்கு உட்பட்டுப் பிழைத்தவற்றினதும் ஈரற் குழியங்களில் கிளைக்கோசேன் காணப்பட்டது. தொழிற்படவிடலுக்கு உட்பட்ட வாற்பேய்களில் ஈரற் குழியங்களில் காணப்பெற்ற கட்டமைப்பு மாற்றங்கள் அப்லாதொட்சினின் நச்சுத்தன்மைக் கலங்களைத் தாக்கியதன் விளைவாக ஏற்பட்டிருக்கலாம்.

---

5. மேற்பரப்பு நீர்ப்பாசன முறைகளுக்கு பயிர்கள் காட்டும் தூண்டற்பேறு.

என். சேனநாயக்கா

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1):

பயிர்த் தூண்டற்பேற்றுடன் தொடர்புடைய நீர்ப்பாசன முறைகளைப் பற்றிய ஒப்பீட்டாய்வொன்று 1973—1974 இல் மேற்கொள்ளப்பட்டது. மதிப்பீடு செய்யப்பெற்ற இரண்டு நீர்பாசன முறைகளுள் நெளிசால் முறையானது சாதாரணமாக வழக்கிலுள்ள உழுசால் முறையினைவிட சிறந்த விளைச்சலைக் கொடுத்தது. ஈரத்திற்கு மிகுந்த தூண்டற்பேறுடைய பயிர்கள் இப்புதிய முறையின் கீழ் அதிக விளைச்சலைக் கொடுத்தன. சாதாரண உழுசால்களைவிட நெளிந்த உழுசால்கள் யாவற்றிலும் ஒரு சீரான விளைச்சல் ஏற்படுவதும் இம்முறையின் இன்னொரு சிறப்பம்சமாகும்.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

6. செம்மறி ஆடுகளுக்கு ஒரே சக்கைத்தின் மூலமாகக் கொடுக்கப்படும் காரப்பண்பூட்டப்பெற்ற வைக்கோல் உணவில் கழிவுத் தேயிலைத் தூள், யூரியா ஆகியவற்றிலுள்ள நைதரசன் உபயோகித்தல்.

எம். சி. என். ஜயசூரியா

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1):

சோடியம் ஐதரொட்சைட்டு கலக்கப்பெற்ற வைக்கோல் உணவில் கழிவுத் தேயிலைத்தூள் (எஸ் ரீ,எல்) உபயோகமாகும் முறையினை ஆய்ந்து அறிதற்குக் கலப்பின செம்மறி ஆடுகளைக் கொண்டு நடத்தப்பெற்ற உணவூட்டப்பரிசோதனை யொன்றின் பெறுபேறுகள் இங்கு தரப்பட்டுள்ளன. அரைத்த சோளத்துடன் (100 கிராம் செறியத்தில் 14 அல்லது 18 கிராம் அளவான) யூரியா அல்லது (100 கிராம் செறியத்தில் 18 கிராம் அளவான) கழிவுத்தேயிலைத் தூள் கலக்கப்பெற்ற மூன்று செறியப்பங்குணவுகள் ஒப்புநோக்கப்பட்டுள்ளன. பண்பூட்டப்பெற்ற வைக்கோல் (4% W/W) ஒரே சக்கைத்தினைக் பகியார்வத்தைத் தூண்டுதற்குக் கொடுக்கப்பட்டது. யூரியா என். பீ. என் உணவுகளை உட்கொண்ட ஆடுகளைப் போன்று கழிவுத் தேயிலைத்தூள் அடங்கிய உணவுகளை ஆடுகள் விரும்பி உண்டன. ஆனால் கழிவுத் தேயிலைத்தூள் கலக்கப்பெற்ற உணவில் 6% முதிராப் புரதப் பொருள் மட்டுமே உள்ளது. கழிவுத் தேயிலைத் தூள் புரதப்பொருள் இடை கடப்புத் தன்மையுடையதாகலாம் என்ற உண்மை இதனால் தெளிவுறும். சோடி யம் ஐதரொட்சைட்டு கலக்கப்பெற்ற வைக்கோல் மூல உணவுகளுக்குக் கழிவுத் தேயிலைத்தூள் சிறந்ததோர் குறைநிரப்புப் பொருளாகவும் அமையலாம்.

---

7. பற்சொத்தைகளிலுள்ள உமிழ் இமியுனைகுலோபியுலின் இலக்ரோபெரின்.

எஸ். திசாநாயக்கா, எல். பீ. சமரநாயக்கா,
ஆர். எம். பென்னற்

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1):

தீவிர பற்சொத்தையுள்ளவர்களிடத்தும் குறைந்த பற்சொத்தையுள்ளவர் களிடத்தும் காணப்படும் சீரம் $^{IG}_{g}$ $^{IA}_{g}$ நிலைகளும் உமிழ் $^{IA}_{g}$ மற்றும் உமிழ் இலக் ரோபெரின் நிலைகளும் கணக்கிடப்பட்டுள்ளன, தீவிர பற்சொத்தை நோயாளி களிடம் இமியுனைகுளோபின், இலக்ரோபெரின் ஆகியவை மிகுந்து காணப் பெற்றன.

---

8. இலங்கையில் சீழுரல் சிரங்கு நோயும் கூர்த்த புன்றிரணை ஊறுநீரியழற்சியும்—எதிர்நோக்குப் பின்நோக்காய்வு.

எஸ். என். அர்சகுலரத்தினம், என். சரவணபவன், சி. நவரத்தினம்,
டீ. ஏ. குணவர்த்தன.

J. Natn. Sci. Coun. Sri Lanka 1981 9 (1):

சீழுரல் தோல் நைவுகளுடன் கூர்த்த புன்றிரணை ஊறுநீரியழற்சி நோயால் மருத்துவசாலையில் அனுமதிக்கப்பெற்ற பல நோயாளிகளை ஆய்ந்தவிடத்து மிகப்பொது (79%) முன்நிகழ் தொற்றுக் காரணியாகச் சீழுரல் சிரங்குநோய் இருப்பது காணப்பட்டது. இந்தக் சுண்டுபிடிப்பின்படி திரிணிடாட், தென் ஆப் பிரிக்கா ஆகிய நாடுகளில் அறிக்கையிடப் பெற்றுள்ளவாறு. சீழுரல் சிரங்கு நோயானது கூர்த்தபுன்றிரணை ஊறுநீரியழற்சி நோய்க்கு பொது முன் நிகழ்க் காரணியாக அமையுமென்பது உறுதியாகிறது. சீழுரல் சிரங்கு நோயால் பீடிக் கப்பெற்ற நோயாளிகளைக் கொண்டு செய்யப்பெற்ற எதிர்நோக்காய்வால் அந்த நோயாளிகளுள் 8 வீதமானோரில் மட்டுமே ஊறுநீர்ப் பிறழ்வுகள் ஏற்பட்டன.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

இந்த வித்தியாசத்திற்கு ஏதுவாகக் கூடிய காரணங்கள் ஆராயப்பட்டுள்ளன. இந்தத் தோல் நைவுகளிலிருந்து தனியாக்கப்பெறற β குருதி இழிசல் இசுற றெப்ரோ கொக்கசுக்களின் சீரவகைக் காட்டுரு ஏனைய நாடுகளில் அறிக்கையிடப் பெறறுள்ள காட்டுருகளோடு ஒத்திருந்தது. கூர்த்த புன்றிரணை ஊறுநீரியழற்சி யால் பீடிக்கப்பெற்ற 40% நோயாளிகளின் தோல் நைவுகளிலிருந்து இசுறறுப் பிலோகொக்கசு அவுரேயுசு மட்டுமே பெறக்கூடியதாய் இருந்ததெனினும் ஊறு நீரியழற்சியை உண்டுபண்ணும் விசேட பற்றீரியாக்கள் இருப்பதனை அவ்வாறு பெறப்பட்ட தனியங்களின் உடமைகள் காட்டுவனவாக அமையவில்லை. கூர்த்த புன்றிரணை ஊறுநீரியழற்சியால் பீடிக்கப்பெற்ற அல்லது பீடிக்கப்பெறுத நோயா ளிகளுடைய தோல் நைவுகளிலிருந்து பெறப்பெற்ற இசுற்றுப்பிலை, அவுரேயுசுத் தனியங்களுள் பொதுப்படையாக தின்குழியத் தொகுதி III அமைந்திருந்தது. அத்துடன் தின்குழிய வகை 54 சிறந்து ஓங்குவதாய் காணப்பட்டது. நம்நாட்டு இசுற்றுப்பிலோகொக்கசுத் தனியங்கள் இடைவெப்ப நாடுகளில் கண்டறிய பெற்றுள்ளவற்றோடு ஒப்புநோக்கப்படுமிடத்து தின்குழியகாட்டுரு பரம்பல் முறை சார்ந்த வித்தியாசங்கள் காணப்பட்டன. தோல்சீழுரலுக்குப் பின் ஏற்படும் கூர்த்த புன்றிரணை ஊறுநீரியழற்சி நோயின் மறைவுக் காலம் ஏனைய நாடுகளில் காணப்படும் காலப்பகுதியை ஒத்திருந்தது; தோற்சீழுரலின் பரப்பு கூர்த்த புன்றிரணை ஊறுநீரியழற்சி நோய் உண்டாவதற்கோ தீவிரமடைதற்கோ தொடர்புடையதாக இருக்கவில்லை. தோற்சீழுரலுக்கு உட்பட்ட ஊறுநீரியழற்சி நோயாளிகளின் ஏ. எஸ். ஓ. நியமனங்கள் சாதாரண நலமுடையோருக்கும் கூர்த்த கீழ்வாதக் காய்ச்சல் கண்டவர்களுக்கும் இடைப்பட்ட நிலையில் இருப் பதாகத் தெரிகிறது. இசுற்றெப்ரோ கொக்கசுக் கூர்த்த புன்றிரணை ஊறுநீரி யழற்சி நோய் சிலருக்கு உண்டான பின்னர் கொள்ளை நோயாகப் பரவிய செய் திகள் இந்நாட்டில் இல்லையெனினும் பகுதிவாரியாகப் பலர் அந்நோயால் அல் லற்படுவதற்கான சான்றுகள் பல உள.

---

9. சிற்றேற்றேகா சீரியலெல்லா என்னும் பூச்சிக்கு வெறுப்பினையும் மயக்கத்தையும் ஊட்டும் உள்ளூர்த் தாவரப் பிழிசாறுகளின் கொதி நீராவி முறை வடியங்களில் உள்ள உடைமைகள் பற்றிய பரிசோதனைக்கூட ஆய்வுகள்.

எஸ். ஆர். கிருஷ்ணராஜா
வீ. கே. கணேஷ்விங்கம்
J. Natn. Sci Coun. Sri Lanka 1981 9 (1):

இலங்கையில் நெற்பயிரைத் தாக்கும் முக்கியதோர் பீடையான சிற்றேற் றேகா சீரியலெல்லா எனப்படும் பூச்சியின் கட்டுப்பாட்டுக்கு உபயோகமான தக வுடைய உள்ளூர்த் தாவரப் பிழிசாறுகளைக் கணிக்கும் பொருட்டு இந்த ஆய்வு மேறகொள்ளப்பட்டது. உபயோகப்பட்ட தாவரப் பொருள் மீது அந்துப் பூச்சி களின் நடத்தையைக் கணிப்பதற்கென "Y" வடிவமுள்ள பூச்சி மனநுகர்ச்சிமானி யொன்று பயன்படுத்தப்பெற்றது. இப்பொருள்களிலிருந்து வெளிவரும் ஆவியின் நச்சுத்தன்மையினைக் கண்டறிதற்கெனப் பெரிய கற்பாத்திரங்களைக் கொண்டு மேலும் பரிசோதனைகள் நடாத்தப்பெற்றன. ஒவ்வொரு பரிசோதனையின் போதும் நேர்படிகள் பல எடுக்கப்பட்டன. பரிசோதனைகளின் போது கிடைத்த தரவுகளைப் புள்ளிவிவரவியல் பகுப்பாய்வுக்கு உட்படுத்தப்பட்டபின்னர், பரி சோதனைகளுக்கெனப் பயன்படுத்தப்பெற்ற பொருள்களுள் விற்றேட்சுநெக் குண்டோ தாவரத்தின் கொதிநீராவியாகும் உட்கூறுகள் நம்பிக்கை தரும் வெறுப் பூட்டியாகச் செயல்படுவதைக் கண்டோம். அத்துடன் சிற்றனெல்லா (எலுமிச் சம் புல்) எண்ணெய் ஆவி சிற்றேற்றேகா சீரியலெல்லா பூச்சிக்குக் கடுமையான நஞ்சாகச் செயல் படுவதையும் கண்டோம்.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

10. இலங்கையில் நைதரசன் உபயோகமும் மாற்றுப் பயிரிடல் சார்ந்த பொருளியல் நலங்களும்.

எச். பி. எம். குணசேனு

J. Natn. Sci Coun. Sri Lanka 1981 9 (1):

இலங்கையில் நைதரசன் உபயோகத்தினையும் மாற்றுப் பயிரிடல், ஒற்றைப் பயிரிடல் முறைமைகளின் பொருளியல் நலன்களையும் ஒப்பு நோக்குதற் பொருட்டு சோளம்/கவ்பீப் பயறு, சோளம்/சோயாப்பயிறு, சோளம்/பச்சைப் பயறு ஆகிய கூட்டுப்பயிர்களைத் துணையாகக் கொண்ட வெளிக்கள ஆய்வுகள் மேற்கொள்ளப் பெற்றன. அவரையமல்லாத பயிர்களுக்கென உள்ளூரில் சிபாரிசு செய்யப்பெற்றுள்ள விகிதங்களில் 0%, 50%, 100% ஆகிய நைதரசன் அளவீடுகள் பயன்படுத்தப் பெற்றன. எல்லாப் பிரதேசங்களிலும் இரண்டு வகைப் பயிரிடல் முறைமைகளின் கீழ் செய்யப்பெற்ற பரிசோதனைகளின் போது அவரையமல்லாத பயிர்கள் நைதரசன் மீது நேர்நிலைப்பட்ட தூண்டற்பேறுடையனவாகக் காணப்பெற்றன. அதிக நைதரசன் அளவுகள் உபயோகிக்கப்பெற்ற நிலைகளின் கீழ் மாற்றுப்பயிராக நாட்டப்பெற்ற அவரையங்களின் விளைச்சல் பாதிக்கப்பட்டிருந்தது. அவரையமல்லாத பயிர்களின் நிழற்படுதல் இப்பாதிப்புக்குக் காரணமாகலாம். மாற்றுப் பயிரிடலால் மொத்தப் பெறுகையும் முழு விளைச்சலும் அதிகரித்தன. மாற்றுப் பயிரிடல் முறைகளின் கீழ் புரத உபயோகத்தின் தேறிய பெறுமானமும் அதிகரித்திருந்தமையால் அத்தகைய பயிர்முறைகள் வளர்முக நாடுகளில் உணவுதியக் கமக்காரர்களுக்குச் சாலச் சிறந்தனவாக அமையுமென்பது வெள்ளிடைமலை.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media

CONTENTS OF PREVIOUS ISSUE

Vol. 8 No. 1 June 1980

Vol. 8 No. 2 December 1980

Short Communication

Appropriate...Te...nology Services
121, POINT... ROAD
NALLUR, ... NA
No. ...
Digitized by Noolaham Foundation
noolaham.org | noolaham.media

Journal of the National Science Council of Sri Lanka

Vol. 9 No. 1 June 1981

Contents

Published by
The National Science Council of Sri Lanka, and
Printed at Swadeshi Printers, 341, Olcott Mawatha, Colombo – 10.

Digitized by Noolaham Foundation.
noolaham.org | noolaham.media