

November 13, 1963.

Dear Dr. Parpia,

I am sending you herewith a note conveying my observations ~~bearing~~ on some of the aspects covered by the report on protein-rich foods, which is now being finalised. I have also made certain suggestions in respect of further work and also indicated certain possible new lines of approach. I have prepared the material primarily with the object of evoking some discussion and further thinking, on the part of our colleagues. It is for you and the others concerned to decide as to what should be done.

Some of the aspects that I have touched on may be of limited interest and may not even be much importance. Some may not require further pursuit. There may, at the same time, be some which may open fresh possibilities and provide scope for intense study for the next decade or more. Sooner or later, such studies may also be taken up in other parts of the world.

In my note, I did not refer to possible future researches on proteins from grasses and other leafy materials. This is an important subject and future success in that field will depend not only on the right choice of material but also on its processing so as to render it acceptable. The R & D Organization of the Defence Ministry has decided to pass on to us the pilot plant which is now with the local Defence Food Laboratory. It is packed and ready and the Institute will have to take it over and instal it in a suitable place. There will also be need to organise a small team that will be exclusively concerned with this line of work. It may well form a part of the joint programme of the 'P' and 'D' Divisions. B&N Division may also be interested, in some of the aspects.

There is ample evidence to show that some of the leaf proteins, singly and in blends, have a high PER comparable with that of milk protein. This is readily understandable when we also consider that the entire production of beef, mutton and milk in countries like New Zealand are based on only two grasses - viz. rye grass and clover. The animals which make marvellous growth derive their entire sustenance exclusively from these two grasses - and without any supplementation with concentrates, minerals etc. as they have in other countries.

whole
It would be hardly necessary for me to emphasise the importance of the subject. The Institute has today some status in the field of protein-rich foods. The future will depend to a large extent, on the maintenance of the lead that we have built up. We should consolidate what we have done and take some of the findings to the stage of practical application both in our country and elsewhere. At the same time, we have to think of and pursue new lines which will lead to fresh development.

As Dr. Schaefer will be visiting us shortly, I would suggest that there may be discussions with different groups to assess the present programme and to consider future programmes of both short and long-range nature.

Although, I am no longer the Principal investigator, I have taken the trouble to prepare the note because of my interest in the subject. I hope you will appreciate the underlying object.

With kind regards,

Sincerely yours,

V. Subrahmanyam
(V. SUBRAHMANYAN)

Scabies

Project on "protein-rich foods"
Report for the year October 1962 to October 1963

A note by Dr.V. Subrahmanyam

During the year under review, several formulations based on blends of vegetable proteins were prepared and evaluated. We should now make a selection out of these and prepare fair quantities for field trials ^{by some of the} ~~colleagues~~ ^{the Venore Group and also other} in India. The first consideration should be given to foods which are suitable for pre-school age children. We should also ascertain the consumer response and the possible interest of the industry in the production of such classes of products.

Side by side with the above, we may explore the possible interest of the UNICEF or other organization in having the products tried out in selected areas. This should be particularly easy in the case of spray-dried formulations, ^{of} which we are now in a position to make substantial quantities with the spray-drier donated to us by the UNICEF.

Our programme should if possible, also link up with that of the I.C.N.^{N.N.D.}~~W.B.~~ especially in countries where diet surveys have already been carried out. We can make and supply the required quantities to selected countries where the ^{existing} national food and nutrition organizations can carry out studies to assess the beneficial effects of the products. Preference should be given to countries which have the raw materials, so that, if they are satisfied, they can take up the production of such products either under their own auspices or through the medium of the industry. The P.A.G. can also associated with such programmes or alternately, we can work through one International agency like the UNICEF. When I met ~~the~~ Mr. Maurice Pate, some months ago, at Edinburgh, he expressed his keen personal interest and also that of the UNICEF in the pursuit of such programmes.

We have not worked out the costs in several of the cases. This must be taken up after ascertaining the acceptability of the products.

For ^{the preparation of} many of the products based on protein isolates it may not always be necessary to go through the integrated process. The meal after ^{expulsion} extraction of oil may be directly extracted according to the procedure worked out by us and the wet slurry of the precipitated protein suitably blended (after adjustment of pH) and then used for spray-drying. For some of the

formulations, roller drying can also be tried, though, owing to lack of equipment, with the required capacity, we may not be in a position to handle more than a few kg. per day. The residue after extraction can be dried or otherwise processed for use as animal feed. The last part will require some intense study.

We have noted that even trituration with water followed by cloth filtration can bring out a fair part of the protein, and the associated carbohydrates from peanut. Similar extraction may also be possible in other cases. Such preparations will reduce the cost and the blends, after cooking, can be used for roller drying.

Quite a large section of people - especially the low income groups - cannot afford to use the processed products exclusively for the feeding of children. They will normally use / them / in addition to the usual feeds, which would normally be diluted forms of the food which the adults consume. With such a background, it should be our endeavour to study the effect of feeding limited quantities, not exceeding 100 grams per day. The supplementary effect should be studied at these levels irrespective of the other types of food received by the children.

2. An interesting thing that has emerged from some of the experiments is that though the PER of the protein in the blend is lower than that of milk, the foods, when fed as supplements to poor cereal diets have given better growth response than with milk foods. The significance of this would require elucidation. A part of the explanation may lie in the beneficial effect of the associated minerals. There may also be some growth factors. It has been reported by earlier workers that coconut water, as also Neera from palm, contain such factors.

3. It has been noted that addition of sucrose, fruit pulp etc improve the growth response to some of the formulations. These will require further confirmation and more extended study to determine the mode of action.

4. Several of our studies, as also those of other colleagues have shown that lactose of milk is a major interfering factor in control of diarrhoea in case of kwashiorkor. This has also often been the complaint which has been received from time to time to t in respect of baby foods based on milk. If there is any infection leading to diarrhoea, the presence of lactose usually aggravates it. One method of dealing with this problem will be to subject the milk to lactic formulation and then to re-adjust the reaction prior to drying by one of the methods. The possible use of such a milk product, with added sucrose and fruit pulp or even pectin for the treatment of kwashiorkor will be of much interest and value. Pec-

tin is already being used for treatment of diarrhoea and other interfering ^{in ~~ingestions~~ ^{in ~~ingestions~~ ^{in ~~ingestions~~}} ingestions. It may be reasonably expected that such a formulation will give a better response than skim milk powder alone.}

The possible extension of the above treatment to some of our formulations containing small percentages of milk powder will also be of value.

5. In South India, every age group above that of the babies, which are nursed by the mothers, receive directly or indirectly, certain food additives like tamarind, chilli, salt, spices etc. The effect of these on protein utilisation has not yet been studied. We have already noted from other studies that in the case of both experimental animals and human subjects such additives make the foods more appetising and lead to distinct increase in food intake (by about 50%) with even greater improvement in growth response. This is an important aspect which may have a bearing on quicker relief of general protein malnutrition and even in the treatment of kwashiorkor. It is well known that kwashiorkor cases have very poor secretions of digestive juices. Such additives may help to stimulate the secretions and even lead to better ^{response} than is now possible. The types of additives to be thus incorporated may be first determined on the basis of acceptable taste by the Dietetic Division and their usefulness verified by animal experiments before trying on children.

This line also offers scope for a large amount of basic work, with evaluation of the function of each component as also the active substances associated with it (eg. tartaric acid, tartarates and pectin in tamarind, capsaicin in the case of chilli, essential oils in the case of spices, curcumin in the case of tumeric etc). Such additives may also have a bearing on the utilisation of other food constituents such as starch, fat etc. Even in kwashiorkor cases, addition of some percentage of fat has been found to be well tolerated and utilised.

6. In all our formulations used for animal experiments, as also trials with human subjects, we have been using salt mixtures, which are, more or less, adaptations based on those originally evolved by Osborne and Mendel. No systematic study has yet been carried out to determine the effect of higher or lower levels of some of the materials. The significance of such a study becomes important when it is realised that some of these elements may have also their effect on the utilisation and response to the protein. During recent years, zinc, potassium and magnesium have assumed considerable significance and their role in protein utilisation may be more important than has yet been realised. There have been reports of zinc deficiency in some parts of the world (eg. Egypt) while the importance of that element in the utilisation of soya protein has been shown by the American workers.

There is a large amount of basic work to be done in ^{the} field in relation to individual proteins, and systematic pursuit of this subject will be highly rewarding.

7. During the early forties some of us had an opportunity to carry out studies in the gastric secretions of under-nourished children. According to Dr. Doraiswamy, such children have very low secretions of acid, rennin and pepsin, thus showing that they have a low capacity for digesting protein. Their bile of secretions are also found to be quite inadequate. The significance of this in relation to protein malnutrition and subsequent improvement through adequate ingestion of protein will provide a very useful ^{line} of study.

We have also observed that the aqueous extract of stomach mucosa contains substance which helps to control oedema through improved excretion of urine. As the ingestion of proteins helps to control oedema, the significance of this in relation to urinary discharge and the nature of the products that are excreted will be of interest and value.

8. Some valuable work has already been done on the nature of urinary metabolites associated with protein malnutrition. It is also known that while improving the condition, different proteins yields their characteristic products of excretion. More recently, there was a report that sweet potato, for instance, invariably yields hippuric acid in human urine. The Hyderabad workers have reported that there is a distinct difference between urinary excretion ^{from} for vegetable proteins and that from animal proteins. Whether each vegetable protein yields its own characteristic products and whether the latter have any bearing on health would be a subject for study. At the same time, we have to appreciate the fact that even vegetarian diets provide proteins from different sources and that we generally have to deal with a mixture rather than proteins from any single source.

A large amount of work has already been done on the influence of the nature of the protein on the composition of the vital organs. This line may not be of immediate interest as we rarely have a single protein as a source of food; though this may be experimented ^{even} under conditions of emergency or acute shortage.

9. None of the studies so far carried out on protein malnutrition have completely ruled out intestinal infection as a related factor. In India, and also in many other countries, the infection - bacterial, protozoal and even worms - may be derived from several sources which may include water, food and insanitary environment. The infection may be repeated and sustained and its effect may be naturally expected to be super-imposed on that of the dietary proteins. It has been reported by some earlier workers that even milk has not produced any beneficial effect in certain areas. We had our own problem in the Chitteri village where the children did not show adequate response

It is difficult to state whether the effect of superimposition of infection of one kind or another can be revealed by rat experiments, though this can be tried. At the same time, there can be a systematic study to determine the nature of the prevalent types of infection in some selected areas by systematic examination of stools and through other observations; to adopt control measures and to study the subsequent effect on the response of the children to protein-rich foods. It is quite conceivable that in presence of certain types of infections, the administration of protein may be even injurious. A good example of this will be the effect of E.Coli, B.typhosus or cholera vilrio all of which are present in many areas - though not always in a virulent form - and which would be favoured by extra protein in the diet. There are also other organisms causing diarrhoea and dysentary and which are persistent and cannot be easily eliminated. Amoebic infections are also frequently present.

The above study will be of long-range importance in relation to the interpretation of response to protein-rich foods in general. It is well known that in many of the Eastern countries, the growth response has no direct relation to the food intake/also to the quality of food. The incipient infection may be a dominating factor in many areas.

It is not clear whether a study of this type can be carried out in association with the vellore group; whether they still will have the staff and facilities for such work. An alternative will be to carry out the study in collaboration with the Indian Institute of Biochemistry and Experimental Medicine which is interested in the control of infections. Some feeding experiments can also be carried out in and around Calcutta where intestinal infections are very common

13 The role of vegetables in the utilisation of protein will also be an important subject for study. Practically every Indian diet includes the some vegetable or other and the nature and quantity of the material consumed depends on the season and the economic status of the family. We should take some prominent types which are also known to be consumed to the largest extent and study their influence on the utilisation of comparatively pure protein in the two selected types of first instance - eg. casein and isolate from peanut.

(P....6)

Vegetables - especially those which come from ^{plants} ~~plants~~, which do not grow tall - are among the chief carriers of infection and this factor may be eliminated by proper cooking before addition to food.

10 In the Vellore _x experiments on the treatment of kwashiorkor, in the hospital, ~~shown~~ about 25% of the cases are reported to have died in spite of high-protein therapy. They were all advanced cases and some of them are also stated to have ^{had} secondary infection which could not be controlled. The ultimate cause of death seems to have been hypoglycemia. This condition was noticeable even before administration of protein which also exercises hypoglycemic action. A possible explanation is that, in the ^{initial} ~~terrierical~~ phase, there is a failure of the digestive functions.

It is for consideration whether under such conditions, administration of pepsin along with Tata-diastase or some such mixtures of enzymes will be of some help. The object of any form of therapy is to minimise the number of casualties even under the worst of conditions. The experiment may be first tried in Mysore and if the results are encouraging, we may contact Vellore or Hyderabad for further studies.

11 The experiments carried out at Walajapet Ashram brought out a striking difference between the two groups of pre-school age children - one on the usual South Indian rice diet and the other on the same diet supplemented with the protein isolate. In addition to showing better growth of response, the children receiving the protein supplement have shown much more of liveliness, both mentally and physically, and this is clearly noticeable even to a superficial observer.

We have already planned for some psychological studies with these children. This line of work should be further pursued and the studies carried out do determine the mode of action of the protein in producing such an effect. There is evidently some hormonal disturbance resulting from protein deficiency and this will require careful study.

12 The results ~~of this~~ so far obtained both by us and other colleagues would show that the protein isolate from peanut is well absorbed and utilised by cases of kwashiorkor. The results seem to be distinctly more favourable than with an equivalent quantity of the edible quality of peanut meal.

While we have not so far found any significant difference between the PER of the protein isolate and that of the protein in the meal, lower results for the former have been invariably reported by Dr. György, though they get nearly equalled on supplementation with vitamin B₁₂.

Although rat experiments have an important place in the evaluation of proteins, we have yet to come to some definite conclusion as to whether the human subjects responds better to the isolates than to the protein in the meal. We have already started some experiments to elucidate this point, but more quantitative studies will be needed ^{before we} to confirm ~~this point~~. ^{Can come to some definite conclusion. If the isolate is found to be superior} After confirmation, we should also know why the protein in the meal is not so well utilised as that ^{of} in the isolate.

The extension of the above observations to soya, cocoanut, sesame and cotton-seed proteins will also be of interest and value. This will be particularly so in the case of cocoanut, the meal from which contains a high percentage of fibre. Rats can consume and utilise even uncooked cocoanut meal ^{at} and fairly high levels, whereas it is very doubtful whether human subjects - especially malnourished children - can. This will require a well planned programme so as to draw some firm conclusion.

13 There has recently been some divergence of opinion in respect of the ^{relation} selection of calcium to protein. In ^{none} more of our experiments we could get adequate response to protein without supplementation with calcium. While our ^{arguments} arrangements have so far, been in favour of calcium supplementation to make up for the ^{well known} deficiency in the diets, we have yet no concrete evidence to show whether the protein itself is better utilised in presence of calcium. This may be particularly of interest in the case of whole or defatted soya flour.

Some carefully planned metabolism studies will help to elucidate this point.

14 The present programme has so far been largely confined to oilseed meals and the isolates derived therefrom.

often in the place of vegetables

Before long, there will be need to have a small team working exclusively on pulses. The Indian experience with pulses has been divergent depending on the region. In some areas (eg. Maharashtra) substantial quantities of different pulses are being regularly consumed ^{without apparently} any adverse effect. On the other hand, ~~the~~ South Indians do not seem to tolerate more than minimum quantities of pulses - and that too of only of certain types. Apart from the possible difference in ~~and~~ adaptation, the nature of the diets and the manner in which they are consumed may probably make a difference.

In Maharashtra, for instance, rice forms only a small part of the diet while the bulk is made up of processed millets and wheat which are made into Chapati which has to be masticated. The South Indians, who are predominantly rice eaters, do not generally masticate the food. This may make a difference in regard to the digestion of pulses which are not often fully cooked and require some mastication.

15 The protein isolate as prepared out of oilseeds by any of the wet methods always contains a small percentage of oil, which may be

of the order of 2 - 3 per cent and sometimes even 5 per cent. This will also include varying proportions of free fatty acid, depending on the initial content of the starting material. Some of the batches have kept very well, while others have shown a tendency to turn rancid. Processed foods incorporating protein with pre-cooked ^{starch} or dextrimaltose have kept well. At the same time, the prevention of the off-flavour and other undesirable consequences in the quality of the isolate itself is very important.

Anti-oxidants seem to have only limited scope. Starch has a protective action and may be effective if it can form a complete protective coat around the ^{particles of} oil or fatty acid. The possibility of incorporating some percentage of pre-cooked starch ~~for~~ along with wet protein (before drying) may be of interest provided lumping can be avoided through quick roller or spray drying. Failing this, a known alternative procedure will be to solvent extract the dry protein, but that will add to the cost. For many of the food products, the wet protein ~~w~~ itself can be used as a part of the blend prior to drying.

Another possible method would be the use of ~~m~~ lime or calcium succrate as a solvent for the wet protein before final precipitation. This will result in the elimination of the fatty acids and also some of the neutral oil in the unextracted residue. Subsequent acidification with HCl with or without the addition of sulphite may yield a nearly fat free protein. This may be worth some systematic study. Removal of free fatty acid may be an essential step for controlling oxidative rancidity.

In the case of cottonseed kernel, it is the rupturing of the cells that is a major contributory factor to the release and reaction of gossypol with the protein and particularly with lysine. The possibility of the extraction from a whole kernel in presence of SO_2 or other reducing agent may offer a possible line of approach. Calcium succrate may again be worth trying.

After extraction of as much protein as possible, the remaining part may be pressed for oil or otherwise processed.

16 The above technique may also be worth trying in the case of soya-bean to determine whether a fair part of the protein can be extracted without affecting the oil or introducing the anti-^{trypsin} enzyme and anti-growth factors. By such ^a treatment we can also eliminate any possible infection during extraction.

17 In the case of fresh coconut, only the technique of maceration in aqueous medium followed by pressing has so far proved useful because we cannot afford to lose the non-protein constituents ^{which} carry to the flavour. As there is however some retention of protein in the residue, a technique of extraction along lines similar to the above may lead to some further recovery while leaving the oil behind.

18 The trials so far carried out on the use of ammonia as a preservative for fish have revealed certain practical possibilities, but the ~~effective~~ effective use of the technique has yet to be demonstrated. By reducing the time of steeping and subsequent packing in closed containers, ^{along with some absorbent material} the loss through dripping may be reduced to a minimum. While using the material for preparation of fish protein concentrate or even for drying with or without added salt, the most attractive form of approach will be to ^{reconstitute} ~~utilise~~ the fish for ^{so as to} table use. It should not be difficult to add back the fish flavour which ^{is} ~~may~~ lost during the process of leaching for removal of ammonia.

~~During the current season, The trials~~ ^{which} are being repeated on a bigger scale. ^{carried out during the current season may throw further light on the possibilities.}

19 The future of fish protein concentrate depends to a large extent on i) successful storage of fish of minimum cost ii) simplification of the technique for extraction of oil and fish odour and preparation of a white, attractive product and iii) retention of the nutritive value. As for i) a treatment like the one with ammonia or some technique of silaging seems to offer the best scope. The method adopted for the production of fish cheese may also ^{provide} ~~allow some~~ means of keeping the fish solids from spoilage. The technique of removal of oil and odour is still the ^{most} elaborate and expensive part. It is here that some fresh approach is needed. The nutritive value ^{appears to be} ~~does~~ ^{affected to some extent} ~~suffer~~ through repeated ^{solvent} extraction. The fish protein also becomes quite insoluble, but that happens even on mere drying.

^{like Bombay Ducks,} A ~~simplified~~ procedure which is worth trying is to start with a fish which is low in fat content and to macerate and cook in a ^{fairly} ~~family~~ acid medium. The resulting curd can also be repeatedly washed with dilute acid which will help to ^{remove} ~~extract~~ only the odouriferous constituents. It is difficult to say whether this process can ever be complete, though it is known to be feasible in the case of shark flesh. Subsequent to this, even a single extraction with a solvent may be sufficient to remove the oil and the residual odour.

In the case of fish material subjected to such treatment, the use of lime or calcium succrate may also provide a means of extracting the protein while leaving the oil and free fatty acids behind.

20 The vellore experiment on the use of alcohol extracted fish flour from sardine for treatment of kwashiorkor yielded a disappointing result. ^{It appears to be a case of the fish protein in that condition not being digested by the ^{grossly malnourished young} children.} The experiments now planned with a roller dried product based on fresh ^{fish material} ~~fish material~~ will reveal whether better results can be obtained with such a material which has been subjected to minimum amount of processing.

21 The reduced food intake and low ~~groups~~ growth response observed in the case of fish flour obtained by solvent extraction of dry fish material which had gone rancid during prolonged storage will be a subject for ^{fruitful} research. This will have a bearing not only on fish flour but

also a variety of other protein rich materials.

Arising from previous discussions, it had been suggested that the oxidation^{ve} rancidity can be catalysed in presence of proteins with known PER. The products can then be extracted for removal of associated fat and then tested for not only the PER but also the intake, ^{and corresponding growth response, examination of vital organs etc.} as compared with the untreated material. This will help to show whether this oxidation^e rancidity causes some permanent damage to the quality of the food - either through direct interaction with the protein ^{or} through leaving some harmful residue which could not be removed by the solvent.

22 The use of heptane for removing moisture by azeotropic distillation and subsequent extraction of oil has been experimentally tried in the case of both fish and coconut.

In the case of coconut, it would be important to work out the material balance and to have some idea of the cost. The results so far obtained are interesting and deserve to be pursued to the logical conclusion.

In the case of fish, while the resulting product is attractive, the protein value is adversely affected. The trials should be repeated and if the findings are again the same, the cause of such effect should be determined.

23 ^{note} The trend of evidence in respect of the effect of protein ^(the anationation of) in diabetes would suggest that there are two distinct phenomena which may or may not be traceable to the same factors. One is the general effect of the protein which helps to improve the physical well-being of the subject. The second one which is more elusive is the effect on carbohydrate metabolism as reflected in the blood sugar level. This action seems to be reasonably consistent only in the case of some proteins like casein when administered as calcium caseinate.

Doubling the dosage of caseinate has not conferred any corresponding benefit. If the reduction of blood sugar level was due to the action of the protein or any of its products of hydrolysis or some mineral or unknown constituent which is carried with the casein, the effect should have been improved through doubling the dosage.

Further development in the line will depend to a large extent on the elucidation of the mode of action of the caseinate. Its effect is certainly pretty quick when ingested with glucose. It is not clear whether the examination of stomach contents at intervals will reveal ^{any} thing, but it is worth trying.

A more interesting approach will be to study the effect of some ^{supplements} ~~additives~~ such as minerals and vitamins, both ^{possibly} ~~slightly~~ and in combination ^{along with the protein} to see whether they can, in any way, influence the action of ^{the} protein. Among the minerals zinc as cation and phosphate as an anion will be of interest. Among the vitamins, thiamine by itself or with some added magnesium may be of ~~some~~ ^{interest. significance.}

24 Other evidence with weight reducing formulations ^{which involve} would suggest that i) reduction of the total intake of carbohydrate and fat and ii) incorporation of some components for reducing appetite (without any injurious effect) would suggest that a fairly good intake (about 70 gms.) of a good class protein like casein with adequate complement of minerals and vitamins would be an useful line of approach. Such a composite diet can also be combined with ad lib intake of certain classes of vegetables (like knol-kol which has been found to be beneficial) and citrus fruits. These may be filling and at the same time, help to reduce the intake of starchy foods and fat.

Polyphenols such as those present in jambulana, myro^blolanⁿ, Indian gooseberry etc. have been ~~reported~~ ^{found} to be useful. Our colleagues in Pakistan are pursuing an intense programme on the utilisation of the active principles present in better gaurd. These can all be incorporated in a dietary regimen and the combined effects studied.

Our efforts have so far been in the direction of the children of the effect of ^{the} protein with an ad libitum feed in respect of the other food constituents, at the choice of the consumer. This approach has been useful in a fair number of early cases, but the more advanced ones have shown only a limited response. As the ultimate object is to control the metabolic disorder even in advanced cases, there should be a concerted approach ^{along some of the above lines} ~~with various additives to provide the bulk but to reduce the intake of carbohydrates and fat.~~ ^{study the over-all response}

No single substance administered orally may be completely effective.

We are today in a position to make weight-reducing formulations which nearly conform to the more expensive ones on the market. As we also know their composition, we should be in a position to try them as formulations for control of diabetes with other additions as outlined above.

25 Of all the food constituents, the one that seems to be more concerned with serious metabolic disturbances - at any rate in the advanced condition - is the fat which leads to the formation of ketone bodies. As the protein is known to have a beneficial effect on the utilisation of fat, a study of the attendant metabolism following the ingestion of a high dosage of protein in the case of a ^{also} diabetic subject would of interest (p. 12)

and value. Vitamin B₁ also plays a definitely favourable role, but that is presumably associated with the carbohydrate metabolism. Its relation to fat between metabolism will also be of interest.

26 The significance of the phosphate rad^{ic}le, by itself, and in combination with protein in the metabolism of a diabetic subject would be of some interest. It has been reported that the plasma inorganic phosphorous in the normal subject tend to increase while that of the diabetic, especially after ingestion of casein, tends to diminish. ~~This observation combined with the known phenomenon by the diabetic in advanced condition, it may be fruitful to study the effect of the ingestion of phosphate with a suitable cation (calcium or potassium) will be of interest.~~ *As the diabetic in the advanced condition is known to excrete phosphate of interest*

27 The ~~improved modification~~ ^{modified procedure} for the estimation of aflatoxin with the use of i) calcium chloride for extraction and ii) thioglycollic acid for the quenching of fluorescence should be tried out more extensively both by us and in collaboration with R.R.C. Hyderabad who are not ¹⁶ engaged on a systematic survey of the groundnut and cake produced in different areas for the incidence of this ^e toxin and its possible association with the local conditions.

28 A highly significant observation is the formation of a fluorescent substance as the result of mere wetting of kernel even in the absence of fungus ^l infection. It is possible that this substance is only an artefact, but unless it is conclusively established that such is the case, we cannot be certain about our ground. This fluorescent substance is ~~found~~ ^{formed} without any discolouration the kernel. It is not formed during the wetting of soya-bean which is also known to form the toxin as the result of fungus growth. ~~The study of this problem deserves high priority, preferably in collaboration with our colleagues of the Tropical Products Institute, London who have done pioneering work on the subject.~~ ^{Further}

Some concrete suggestions/for implementing the above proposal have already been made.

The product ^{normal} formed during the wetting of groundnut kernel should be regarded as a metabolite because it should inevitably be formed during the germination of groundnut. It is presumably ~~metabolised~~ ^{utilised or otherwise transferred} by the groundnut seedling. This can be easily verified even in the laboratory by conducting some germination studies. ~~In that case,~~ ^{transferred into the kernel} it is also obvious that it is not ~~reformed~~ during the formation of groundnut after the maturation of the plant. This line of study will also be of interest to Dr. Altschul and his associates of the pioneering Research unit of the Southern Regional Laboratory at New Orleans. From our end, we may plan some

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Even if it remains unchanged in the groundnut plant

histo-chemical studies to trace the fate of the fluorescent substance in the groundnut seedling.

It may be of interest to mention that in several parts of Mysore (I have recently seen several large areas in Chamara-nagar Taluk) ^{the groundnut crops now on the field} ~~which are more under groundnut.~~ There may be some areas even in the close vicinity of Mysore. It may be of interest to examine the growing plant and the different parts for the possible presence of the fluorescent substance. If, by any chance, it is ^{and continues to exist in different parts} normal metabolite of the plant, ~~the whole picture, will~~ may assume a different pattern.

A further aspect which would be of practical significance is that the method of treatment for controlling infection will have to be modified if it turns out that the fluorescent substance formed as the result of wetting the kernel is itself toxic or in ^s a precursor ^{to} the formation of the toxin by the fungus. As the formation of that substance and fungus growth are both favoured ^{by me} a high moisture content of ~~the~~ groundnut, quick drying of groundnut in shell would ^{then} be the most effective practical solution.

29 Independent of the above considerations, we should extend our studies on the control of fungus growth on groundnut which could not be dried easily consequent on inclement weather and other unfavourable conditions. Ammonia applied as such or generated in situ by combination of lime and ammonium sulphate is effective, but ^{or} ~~this~~ is a suggestion that lime itself ^{may} ~~can~~ be useful if it can completely cover the shell. There are also ~~of~~ other possible treatments, including ^{the} use of fumigants and certain chemicals. If one or more of such treatments are effective, we can certainly check the growth of fungus and the attendant formation of aflatoxin. There ^r will also be ^a larger recovery of the kernel than is now possible under such conditions. The market value of the product will also increase. Any treatment that is recommended should be based on materials ^{which are} easily available in the country and at the same time, cheap and non-hazardous.

After completion of the laboratory studies, we should confirm the findings under field conditions and then pass on the results to the Government and to the Indian Central Oilseeds Committee, ^{for being} ~~having~~ followed up through various concerned agencies.

January 21, 1964

Dear Dr. Parpia:

I am finding it difficult to fill up all the columns of the 'Research Project Proforma' for want of information about certain aspects. I can, therefore, mention certain of my requirements only in general terms:

Name of subject/Project:

1. Influence of certain additives on utilisation of food constituents and growth response - with particular reference to proteins
2. Effect of proteins and minerals at different levels on growth rate and distribution of body weight.

There will also be other studies on which preliminary work will be carried out before planning for extended programmes.

Short range objectives of the Projects:

1. There has not so far been any systematic work on the influence of appetisers, as commodity used in our dietary spices and flavouring substances, sugars, pectins, certain minerals, vitamins (especially B₁₂) on the absorption and utilisation of food constituents. The experiments will be first carried out with albino rats with metabolism and other related studies. Initially, synthetic diets will be tried, but will be followed later by some of the diet compositions as commonly used.
2. The influence of the nature and level of protein on body weight and the manner of its distribution will also be studied with albino rats in the first instance. Synthetic diets with constant percentages of fat and other constituents will be first tried. The influence of some of the factors listed under (1) will be studied later.

Side by side with the above, some preliminary studies will also be carried out on 1. the influence of

certain ~~ixstix~~ factors such as temperature and certain filters on the extraction of proteins, in ~~fxz~~ fat-free condition, from biological materials associated with fat and flavouring substances; 2. effect of certain pre-treatments on some vegetable proteins (especially glabulins) to facilitate their extended linkage to form complex molecules or to denature after mild processing and generally to improve their binding property; 3. Study of certain factors controlling the respiration rates of tropical fruits and vegetables and 4. determination of critical moisture level at which certain flavour constituents are lost from some food materials.

There may also be other related studies. The technique of wet digestion which was developed some years ago will be applied for nitrogen metabolism studies.

Long range objectives: Every one of the above listed subjects is expected to offer scope for practical application. The immediate object is to carry out the work as fundamental studies and then to leave it to others to follow up. Some new programmes may also develop from the above studies, but they cannot be forecast at the present time.

Date of starting: By about the middle of February 1964 if space, staff and facilities are available.

Target date for completion: The immediate programmes may take about 2 years, but the completed studies may take several more years.

Expenditure: Recurring (Exclusive of salaries):

About Rs. 20,000/- per annum

Capital: Rs. 40,000 will be required for first year.

Personnel: The project will require at least one S.S.O., two S.S.A.'s, one J.S.A., two Lab. Assistants and two attenders to begin with. More staff will be required as the project expands in scope and volume.

In addition to guiding the staff, I will also do some work on my own.

As for the qualifications of the staff, I would require at least two senior people with previous research experience

in Biochemistry and Nutrition, and the others with general (P..3)

Dr. Bhatia, with whom I had a preliminary talk, was kind enough to advise that he can spare some three or four people with the types of qualifications that I require. He also advised that he can spare fair amount of equipment and apparatus to start with. This will be necessary because the Stores may not have much to spare. One fair-sized refrigerator will also be required. I have no doubt that he also had some talk with you about this subject.

My object is primarily to pursue certain concepts of mine and to lay the foundation for any fresh line of development. At the present time, no collaboration with any Division is contemplated, but their assistance in the matter facilities may be required. I may also pass on the findings to the others to pursue, if they are interested.

Some of the programmes that I have outlined are covered by other notes that I have sent to you from time to time.

You have already kindly promised me all required facilities and I would thank you to kindly arrange for them at least by the beginning of next month.

In connection with the engagements that I have already spoken to you about, I will be out of station from the afternoon of 28th till the evening of the 31st. I hope that we will have a chance to meet as soon as possible after my return.

Sincerely yours,

(V. Subrahmanyam)

VS: ksd/

Dr. H.A.B. Parpia,
Director,
CFTRI, Mysore.

True copy

[Handwritten signature]

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Dr. H.A.B. Parpia,
Director,
CFTRI, Mysore.

True copy
[Signature]

November 19, 1963.

Dear Dr. Parpia,

With reference to your note dated November 15, 1963 my immediate, personal interest is largely to concentrate on a single project on which very little work, has, so far, been done and which, I hope, will yield fruitful results. The proposed programme will not also interfere with the interests or claims of any of the other groups. I had referred to certain aspects of it in the note which I had sent under cover of my letter dated November 13, 1963 as addressed to you.

A provisional title for the programme will be 'Effect of different additives on the digestion and utilization of proteins'.

I will like to do the work in collaboration with Dr. Swaminathan and his associates, if both you and he are agreeable to the arrangement.

If the proposal is agreed to in principle, the detailed programme and also the staff required for the work can be gone into at a later date. A small beginning can first be made and the scope gradually expanded after addition of more staff when the vacancies are filled up. I will be guided by Dr. Swaminathan regarding the housing of the staff.

There are some other programmes which I had initiated before you took and I would like to see them completed. They need not take up much of my time. I will list them as follows:

1. Treatment of wet groundnut (in shell) to arrest fungus growth without affecting the quality of the oil or the meal.

This was started x jointly with Dr. Rajagopalan, Shri S.K. Maxjundar and Dr. Sreenivasamurthy. We have already got some promising results. I would like to see this programme completed and the results published. I am not interested in the extension or development which will come under the purview of others.

2. Standardization of the composition of a protein-rich weight-reducing formulation.

This programme was taken up jointly with Dr. Swaminathan and Shri M.R. Chandrasekhara. We have already made a fair amount of kaxdxk headway. Some more remains to be done including the possible use of vegetable protein isolate in place of milk protein. The experimental batches have so far been tried only on a few subjects. More trials and related evaluations will also be needed. After that, I will leave the developmental programme to the others.

3. Studies on the effect of certain treatments bearing on the denaturation of groundnut protein so as to improve the washability and cooking quality of tapioca macaroni and related products.

The above will be essentially a basic work leading to improvement in the binding properties of the protein. The problem is a very important one, but we cannot expect to get quick results. The laboratory trials will be carried out with Dr. Bhatia and Shri Bains as and when fresh ideas occur to us. It will not take much time. Once some conclusive results are obtained, there will then be need for trials with the plant.

4. Standardization of conditions for the ammonia preservation of fish.

Having started this programme, I want to complete it during the next season with Drs. Lahiry and Moorjani. The really important thing is to confirm ^{that} the protein-value is not affected. I will then leave the developmental programme to others.

5. The work on the preservation of arecanut fruit is more or less complete. There are only a few more trials to be carried out. The work will soon be written up for publications. I will leave the extension to Shri Govindarajan and his associates.

There is a little more to be done on the preservation of pea. I would like to see through one more set of trials. After the basic work is completed, I would leave further studies and development to Dr. Srivatsava and his associates. I will also leave studies with other products along similar lines Dr. Srivastava and his group.

~~xxxxx~~ You are the head of the Institute and I do not know whether it would be necessary for you to consult the others in respect of the above. It is for you to take the decision. I do not think that any one will deny my association with those programmes. If you feel that I should sever my connection with those lines, I will gladly do so, provided my past association is not forgotten; and provided I have no responsibility for any subsequent difficulty or failure arising out of such programmes.

I had also started some other lines, but many of them are at a stage when my association is no longer required. They are in safe hands and I hope that they will be carried to their logical conclusion. Both you and the officers concerned are welcome to discuss them with me, if so desired.

My major contribution to the research work of the Institute was through suggestion and pursuit of some new lines of thought. It is, therefore, for you to consider whether you can utilise my association in any other way. My interest will be only in scientific work and, that too, only with your knowledge. I do not, however, wish to be misunderstood if some of the staff consult me about their work, whether they come in the above or any other category. If anything interesting emerges from such discussions, I will gladly keep you informed.

It would perhaps be helpful if we can have some preliminary talk before you take any decision. I am no stranger and my main interest is to go with some work as long as I am able to do so. I have some standing in the profession and I am offering whatever little knowledge and experience that I possess, so that they may be of some use to you and to the Institute.

With regards,

Sincerely yours,

Sd.

(V.Subrahmanyam)

P.S. The date of starting or continuance of any work need not be determined by the period of my leave. I do not propose, for the present, to go out for any long period and as you know already, I have been coming to the Institute on every day, including holidays.

Dr. H.A.AB. Parpia,
Director,
CFTRI,
Mysore.

/ COPY /

Handwritten signature

Copy of the letter dated February 12, 1964 from
Dr. V. Subrahmanyam, Central Food Technological
Research Institute, Mysore-2 addressed to Dr.H.A.B.
Parpia, Director, CFTRI, Mysore-2.

"You may kindly recall that from about the time of the expiry of my refused leave during December, 1963 I have talked to you, from time to time, about the facilities for my research work. You were kind enough to readily agree and to indicate that you had already reserved a room for my work. You also mentioned that you had thought of the staff, equipment and other facilities which could be made available to me. Acting on your advice, I also sent you the required particulars about the programme of research, in which I am interested and which would be of both short and long range importance. I had, in fact, selected subjects which would be complementary to the work that is already being done in the Institute and which I trust, will also lead to some practical application.

As I have not so far heard anything from you, and as I find that you are now busy with the allocation of workers to different disciplines, I would thank you to kindly advise me about the provision that you are making for my work. It is needless to state that I would like to get things started at as early a date as possible.

With kind regards, "

/ COPY /

Handwritten signature

Copy of the letter dated 26 February, 1964 from Dr. H.A.B. Parpia, Director, Central Food Technological Research Institute, Mysore addressed to Dr.V.Subrahmanyam, Emeritus Scientist, CFTRI, Mysore-2.

"I have received your personal note this morning and I am sorry to note that you have misquoted me. I had mentioned to you about two months ago that it may be possible to find some working space, but that I could decide on it only after I had gone through the work of the Institute and studied proper allocation of space for the workers. Therefore, your statement that I had made any promise for additional space is wrong.

Yesterday, I explained to you the entire situation that in order to accommodate our existing staff and provide them normal working facilities, we require at least 50% more space. When this problem was discussed with other colleagues, most of them have converted their office rooms into laboratories or instruments room. I, therefore, again suggest to you that you should utilise your office room for a Laboratory. Actually one Laboratory bench is already fixed in your office which was being used by Major Iyengar when he was occupying that place. If you wish I shall arrange to fix another bench. Beyond this, I regret it is not possible for me to give you any more accommodation for the present. Alternatively, I could approach the C.S.I.R. and seek their permission to arrange for a Laboratory in your house. This may even save you the trouble of coming to the Institute everyday except for Library reference.

I am further disappointed to note that you do not wish to be equated to other younger workers. Even those who are beginning their work have to serve the Country for a long time through acquiring proper experience. I would have normally expected that you would want them to have the best possible facilities in the wider interest of the Country.

With kind regards,"

P.S.:

It is more than four months since you became Emeritus Scientist; if you wanted to do some work, you could have started it by now, as facilities are already available in your room.

/ TRUE COPY /

Y. S. Subrahmanyam