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Research Paper

THE EFFECT OF GUM ARABICON CALF KIDNEY TISSUE CULTURE GROWTH *IN VITRO*

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Abstract

This study was conducted to evaluate the effect of adding different levels of bacterial metabolites supplemented or not supplemented with different levels of Gum Arabic and as well as different amounts of Gum Arabic only on calf and lamb kidney tissue culture *in vitro*. It resulted in more healthy and better growth when comparedwith the control cells. The best result was obtained in the tissue cultures when the G.A. was added to *Klebsiella* spp.and*E. coli*cultures at the levels of 0.5%,0.1% and 1% respectively. The best growth was achieved when an amount of 0.08ml of the*Klebsiella* spp. and*E. coli*metabolites supplemented by G.A. were added followed by 0.04 ml of the*Klebsiella* spp. and*E. coli* metabolites supplemented by G.A..

Key words: Calf kidney, tissue culture, E.coli, Klebsiella spp. metabolites.

INTRODUCTION

Gum Arabic is an exudate collected from the stems and branches of *Acacia senegal*and other related African species of Acacia. GA is a natural polymer consists mainly of high molecular weight polysaccharides and contains high concentrations of calcium, magnesium and potassium salts, which yield arabinose, galactose, rhamnose and glucuronic acid following hydrolysis [1]. Sudan produces about 80% of the world supply of Gum Arabic[2].

The number of end-stage renal disease patients requiring renal replacement therapy RRT has increased dramatically throughout the world during the last decades [3]. High cost of treating patients being a burden on the health care systems of both rich, developed and poor, developing countries, especially the poor ones.Many publications on the impact of nutrition on kidney disease [4], [5].Protein restriction regimen was used in many dietary tries to treat chronic renal failure (CRF) and to decrease uremia [6];[7].Supplementation of fermentable carbohydrate (FC) is a recent approach in the diet of kidney disease patients [8]. This has been claimed to result in a similar urea-lowering effect by increasing urea nitrogen (N)excretion in stools, with a simultaneous decrease in the total N excreted in urine of adults [9]; [10]; [11]and children [12];[13];[14].

Since GA is a soluble fermentable dietary fibre it will increase the number of bacterial mass in the human colon and hence utilization of the urea by the action of bacterial urease, and

elimination of ammonia which is used for the synthesis of non-essential amino acids and protein for human subject and microorganism [15]; [16].Also metabolites of these microorganisms such as volatile fatty acids and polyamines play a major role in regulation of DNA, RNA, Protein synthesis as well as enhancing epithelial cell proliferation [17]; [18].

[19] reported that addition of 10g of GA daily to the drinking water for 4 weeks for man increases the numbers of Bifidobacteria, Lactobacilli and Bacteroides and they were significantly higher for gum arabic than for inulin. It is concluded that gum arabic establishes prebiotic efficacy, at least as good as inulin

[20]mentioned that treatment with Arabic gum (GA) protected the rats fromgentamycine(GM) - induced nephrotoxicity as evident by normalisation of these parameters.G A totally prevented the GM-induced rise in kidney tissue contents of MDA. Kidney histology of the tissue from GM-treated rats showed necrosis and desquamation of tubular epithelial cells in renal cortex as well as interstitial nephritis. Whereas it was very much comparable to control when GA was co-administered with GM. In conclusion, GA protected the rats from GM-induced nephrotoxicity, possibly, at least in part through inhibition of the production of oxygen free radicals that cause lipid peroxidation.

Despite it has become a well-established fact that consumption of Gum Arabic improves the condition of chronic renal failure (CRF) patients by reducing urea excretion via kidneys, the true and real mechanism of doing this is still uncertain. Depending on these facts this study was designed to evaluate the role of Gum Arabic *per se* as well as metabolites of bacteria supplemented with different concentrations of Gum Arabic on the performance of kidney tissue cultureinvitro.(In tissue culture flasks).

MATERIALS AND METHODS

- Calf and lamb kidney tissue.

-*Metabolites of E. coli* and *Klebsiella*spp. cultured in RCM media supplemented with different concentrations of Gum Arabic or without Gum Arabic (Fine powder from Khartoum Dialysis and kidney Transplantation center, its physiochemical properties are mentioned elsewhere [21]were prepared..

Virus: Sheep pox virus (Beaudate strain, international for Sudan obtained from the Central Veterinary Research Laboratories, Soba) was tested for its growth in different batches.

Preparation of bacterial metabolites

E. coli and*Klebsiella*spp. organisms were cultured in Reinforced ClosteridialMedia (RCM) and to another set of bottles of RCM 0.5%, 0.1% and 1% of Gum Arabic were added and then cultured with *E. coli* and*Klebsiella*. Then bottles were incubated at 37°C for 24 hrs. The cultured media were centrifuged. Thefiltrate was sterilized with a 0.2 µm Millipore filter. The solutionswerestored in sterile bottlesfor later use.

Effect of Gum Arabic on cell culture

Extraction of *E. coli* and *Klebsiella*spp. metabolites cultured in RCM media not supplemented or supplemented with different concentrations of Gum Arabic was performed. Then different amounts of these metabolites were added to calf and lamb kidney cell culture. Also Gum Arabic alone in different concentrations were added.

The experimental protocol exhibited as follows:

1- Cell culture with 0.04 ml , 0.08 ml and 0.1 ml *Klebsiela*spp. metabolite supplemented with 0.1%., 0.5% and 1% G.A

2- Cell culture with 0.04 ml, 0.08 ml and 0.1 ml *Klebsiela*spp. metabolite.

3- Cell culture with 0.04 ml, 0.08 ml and 0.1 ml *E.coli* metabolite supplemented with 0.1%., 0.5% and 1% G.A.

4 – Cell culture with 0.04 ml,0.08 ml and 0.1 ml of E.coli metabolite.

5 - Cell culture with 0.04 ml and 0.08 ml of 5.0% Gum Arabic solution.

6- Cell culture only as control.

RESULTS

All batches of tissue cultures showed better growth than the control. The best result was obtained in the tissue cultures when theywere supplied with metabolitesof *E. coli* and *Klebsiella spp.*supplemented with G.A. concentration of 0.5%, 0.1% and 1% respectively. The best growth was obtained when an amount of 0.08ml of the metabolites supplemented by 0.5% G.A. were added. It resulted in more healthy and better growth mainly when the culture media supplemented with Gum Arabic (G.A.) in comparison to control cells (See Fig.1-8).

Fig (1): Calf kidney cell culture provided with 0.04 ml metabolite of *Klebsiella* spp. grown in medium containing Gum Arabic (0.5%)

Fig (2): Calf kidney cell culture provided with 0.08 ml metabolite of *Klebsiella* spp. grown in medium containing Gum Arabic (0.5%)

Fig (3): Calf kidney cell culture provided with 0.04ml metabolite of *Klebsiella* spp.

Fig (4): Calf kidney cell culture provided with 0.04 ml metabolite of *E. coli* grown in medium containing Gum Arabic (0.5%)

Fig (5): Calf kidney cell culture provided with 0.08 ml metabolite of *E. coli* grown in medium containing Gum Arabic (0.5%)

Fig (6): Calf kidney cell culture provided with 0.08ml metabolite of *E. coli*

Fig (7): Calf kidney cell culture provided with 0.04ml **G. A. (0.5 %) only**

Fig (8): Calf kidney cell culture (control)

Also lamb kidney tissue culture gave the same results
Also growth of sheep pox virus was examined and it was improved in all experimental sets.

DISCUSSION

All batches of tissue cultures showed better growth than the control. The best result was obtained in the tissue cultures when they were supplied with metabolites of *E. coli* and *Klebsiellaspp.*supplemented with G.A. concentration of 0.5%, 0.1% and 1% respectively.Such an observation may need more research in solving tissue damage problems as in kidney damage.
The best results were observed in 0.08 ml and 0.04 ml amounts of the bacterial metabolitessupplemented with gum Arabic followed by 0.08 ml and 0.04 ml amounts of the bacterial metabolites only respectively. This may be associated with increased bacterial growth and due to the fact that metabolites of these microorganisms such as volatile fatty acids and

polyamines play a major role in regulation of DNA, protein, synthesis as well as enhancing epithelial cell proliferation. The results of this study validate the findings of [17] and [18] in the last piece of information.It is clear that the addition of G.A. to tissue cultures improve the criteria and the purpose of the application. No similar results were detected in the literature, and this is a first original report of such an observationinvitro. This could give a clue to the observation made by[22] that anurea persons start to pass urine when fed with G.A.

CONCLUSION AND RECOMMENDATION

Metabolites of *E. coli* and *Klebsiella spp.* grown in media with G.A. improved the growth of kidney tissue culture compared to the control. This will give a clue to the fact that anurea persons start to pass urine when G.A. incorporated in their diets.
G.A. can be supplemented to the cell tissue cultures to improve such materials especially for vaccine production.

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