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Short Communication

Involvement of fucose in bacterial oxidative stress processes

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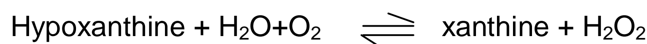
In this research, bacteria oxidative stress processes was investigated in order to evaluate the role of L-fucose correlated with xanthine oxidase activity in the release of toxin generated by pathogenic bacteria such as *Staphylococcus aureus*, *Klebsiella peneumonia* and *Escherichia coli* (display as normal flora). The ratio of total fucose/ total protein obtained proved that L- fucose can act as an antioxidant factor in cell. The obtained ratio increased 4 and 8.37 times fold in *Staphylococcus aureus* and *Klebsiella peneumonia* respectively than in normal bacteria.

Keywords: Bacteria, oxidative stress, L-fucose, xanthine oxidase. (XO)

L-fucose is a 6-deoxy-monosaccharide, widely occurring in glycoconjugates from many different sources (microorganisms, plants and animals); it is formed from GDP-O-mannose via a metabolic pathway in bacteria and animal tissues as reported by Ginsburg (1960). Fucose is one of the eight essential sugars linked with many proteins at the cell surface, which is require for optimal function of cell-to-cell communication (Marks et al., 2004), and can bound to proteins by both N- and O- linked e.g. linked to the Glc NAc residue attached to Asn in the N-linked species (Becker and Lowe, 2003). Xanthine oxidase is a highly versatile and ubiquitous complex molybdopterin, first identified a century ago in milk (Schardinger, 1902). Most of xanthine metabolizing organisms possess a molybdopterin cofactor-containing enzyme that oxidizes xanthine to uric acid (Hille, 2005), and can play a significant role in the detoxification or activation of endogenous compounds and xenobiotics (Borges et al., 2002).



Therefore, bacteria possess an array of detoxification pathways for toxins; by some route antioxidant conjugate with toxin leading to a decrease in the pH of bacterial cell, which protect them against the toxic effects of free radicals (Ferguson et al., 1998).

The aim of the present study is to evaluate the capacity of fucose to act as antioxidant, specifically, a toxin producer in bacteria,

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Materials and Methods

All chemicals and materials used in this study were of high analytical standard

Collection of specimens

The specimens were generally collected from different sites of infection. Blood samples were taken from healthy persons to isolate normal microflora. Mid-stream urine samples were collected from patients with urinary tract infections; all samples were inoculated on the culture media (MacConkey, blood and nutrient agar medium) and incubated aerobically at 37°C for 24-48 h. The bacterial isolates were identified after staining with gram stain; specific biochemical tests were done to reach the final identification according to McFadden (2000).

Estimation of total L-fucose (TF) concentration

This was estimated according to the method illustrated by Dische and Shettles (1948).

Determination of serum protein bound fucose (PBF)

This measurement was determined according to the method illustrated by Dische and Shettles (1948).

Determination of toxin protein (TP)

A kit depending Biuret method was used in the determination of total protein.

Estimation of xanthine oxidase activity

This was done according to the method illustrated by Ackermann and Brill (1974).

Estimation of uric acid concentration

QuantiChrom™ Uric Acid Assay Kit was used in the determination of uric acid concentration

Results and Discussion

The results of this study are presented in Table 1.

Table 1. Variable amounts of biological measurement of the different types of bacteria used in this study.

Test	***E. coli***	***Staph. aureus***	***Klebsiella***
Xanthine oxidase activity using xanthine as substrate (U/L)	11.84	24.75	26
Xanthine oxidase activity using hypoxanthine as substrate(U/L)	0.025	0.031	0.033
Total L-fucose (g/dl)	0.00497	0.0189	0.0327
Uric acid (mg/dl)	2.73	3.55	3.93
Total protein (g/dl)	1.4	1.3	1.1
Protein bound fucose PBF (g/dl)	0.0017	0.000394	0.000228

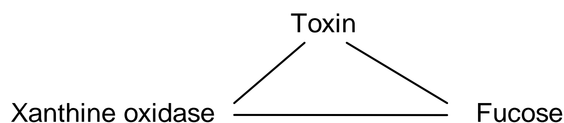
From the results in Table 1 above, xanthine oxidase activity in the three types of bacteria increase in the order of *E. coli* < *S. aureus* < *Klebsiella*, which mean that the pathogenic bacteria has to be more active in order to overcome this violence properties. However, the higher value of xanthine using xanthine as substrate than when using hypoxanthine as substrate is because xanthine binds more rapidly than hypoxanthine. Increased activity was observed in *Klebsiella* than the other types according to following order *Klebsiella* > *S. aureus* > *E. coli* . This is due to the need to overcome the effect of toxins by introducing more uric

acid as a product of the reaction. The amount of uric acid increased proportionally with the activity of XO, therefore, uric acid as an antioxidant was able to maintain the status (that is, its ability to overcome the predominant free radicals). Thus, L-fucose can play an important role to balance oxidative stress status and work as an important antioxidant as shown in Table 1, that is, as long as uric acid increase in concentration, it will increase accordingly to keep the normal status of such media. The ratio of total fucose/total protein (TF/TP) is a determining factor in the activities of fucose as an antioxidant in cells (Table 2).

Table 2. The ratio of TF/TP in broth of different types of bacteria

E. coli	*S. aureus*	*Klebsiella*
0.00355	0.01453	0.02973

The results reflect an elevation in TF/TP ratio in pathogenic species compared with non-pathogenic species. The elevation was seen to be 8.37-times folds in *Klebsiella* and 4-times folds in *S. aureus* than in the non-pathogenic species (*E. coli*). The suggestion for this variation could be due to the high toxins released by the pathogenic species, and a proportionate increase in the defense system leading to an increase in the TF level, that is, severity of oxidative stress cycle (Novaes et al., 1980). On the other hand, another suggestion for this increase could be the breakdown of glycoprotein, that is, xanthine oxidase (a glycoprotein enzyme which has the property of oxidative compound), responsible for the release of free radicals due to its catalytic reaction of converting hypoxanthine and xanthine to urate, therefore, this study concentrated on the effect of both fucose and XO on oxidative stress (antioxidant) as represented by the following scheme:



From the scheme, it is obvious that fucose has synergetic property. It was as a result of the activities of xanthine oxidase that fucose is being release leading to the formation of toxins in pathogenic bacteria (Ali, 2011). Thus, the cell defenses itself by increasing antioxidant marker in which fucose plays a vital role (Govand, 2013). Therefore, L-fucose needs to be syntheses by fucosylation process in bacteria broth medium to enhance cell defense self-system. Table 1 shows that protein bound fucose (PBF) was in low amount in pathogenic species whereas in *E. coli* its amount was 0.0017 g/dl; this gave L-fucose the enabling environment to act as an antioxidant.

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