

**Incorporation of selected herbal extracts
to augment rice with anti-glycation
activity**

By

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rice with anti-glycation activity**

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Declaration of the Candidate

The work described in this thesis was carried out by me under the supervision of Professors S.B. Navaratne and I. Wickramasinghe and this dissertation has not been submitted in whole or in part to any university or any other institution for another Degree/Diploma.

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Abbreviations

ABTS -2,2'-Azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt

AGA-Anti-Glycation Activity

AGE- Advanced Glycation End-product

Al-Aluminum

ANOVA-Analysis of Variance

AOA-Anti-Oxidant Activity

AOAC- Association of Official Analytical Chemists

APC-Aerobic Plate Count

AT- Ambalantota

BG- Bathalegoda

BSA- Bovine Serum Albumin

BW- Bombuwella

CEL- N 3-carboxyethyllysine

CML- N 3-carboxymethyl-lysine

DM- Diabetes Mellitus

DMSO- Dimethyl Sulfoxide

DPPH- 1,1-Diphenyl-2- Picrylhydrazine

EC₅₀-Effective Concentration 50

FAOX-Fructosyl-Amine Oxidase

FN3K-Fructosamine-3-Kinase

FRAP-Ferric ion Reducing Power

FR-Free Radical

GAE- Gallic Acid Equivalent

GK- Normal Green gram treated with *Salacia reticulata*

GLO- Glyoxalase

GM- Normal Green gram treated with *Syzygium cumini*

GMD- Geometric Mean Diameter

GN- Untreated raw green gram

GRP-Glycation Reversing Potential

GSH-Glutathione

IC₅₀-Inhibition Concentration 50

IDDM-Insulin Dependent Diabetes Mellitus

IDF- International Diabetes Federation

IGK- Instant Green gram treated with *Salacia reticulata*

IGM- Instant Green gram treated with *Syzygium cumini*

IRK- Instant Rice treated with *Salacia reticulata*

IRM- Instant Rice treated with *Syzygium cumini*

IRN- Instant Rice Normal (Untreated)

LC₅₀ -Lethal Concentration 50

LD₅₀-Lethal Dosage 50

LDPE- Low-Density Polyethylene

MG-Methylglyoxal

MI- Maha Illupallama

NCDs- Non-Communicable Diseases

NIDDM-Non-Insulin-Dependent Diabetes Mellitus

OECD- Organization for Economic Co-operation and Development

PB-Phosphate Buffer

PBS- Phosphate Buffer Saline

PET-Polyethylene Terephthalate

PRGT- Herbal Treated Porridge mix

PRGUT-Herbal Untreated Porridge mix

QE- Quercetin Equivalent

RAGE- Receptors for Advanced Glycation End-product

RCS-Reactive Carbonyl Species

RN-Untreated normal raw rice

RNS -Reactive Nitrogen Species

ROS-Reactive Oxygen Species

RRK- Raw Rice treated with *Salacia reticulata*

RRM- Raw Rice treated with *Syzygium cumini*

SD- Standard Deviation

SLS-Sri Lankan Standards

T1DM-Type I Diabetes Mellitus

T2DM-Type 2 Diabetes Mellitus

TCA- Trichloroacetic acid

TE- Trolox Equivalent

TFC- Total Flavonoid Content

TMR- Total Milled Recoveries

TPC-Total Polyphenol Content

TPTZ- 2,4,6-Tripyridyl-striazine

WAC-Water Absorption Capacity

WHO-World Health Organization

YMC-Yeast and Mold Count

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ABSTRACT

The excessive formation of advanced glycation end-products via non-enzymatic glycation reactions in human body mediate for much health complication. The scope of this study was to provide a dietary solution by developing anti-glycation activity augmented rice (and related food products) to inhibit the formation of so-called compounds; thereby overcome the emerging health issue. The augmentation of selected rice variety (*Kahamaala*) with anti-glycation activity was carried out by steeping the prepared rice samples (parboiled, raw, and instant forms) in a selected herbal extract (*Syzygium cumini*) consisted of strong anti-glycation activity (IC_{50} :9.75 μ g/ml), in a predetermined time period at 70 °C. Herbal treated green gram (raw and instant forms) was also prepared by following the same method. Subsequently, instant porridge mix was also formulated by incorporating the herbal treated rice and green gram. The effectiveness of the augmentation was assessed by measuring the increment of the (*in vitro*) anti-glycation activity and antioxidant activity (total polyphenol content (TPC), total flavonoid content (TFC), Ferric ion Reducing Power (FRAP), ABTS and DPPH radical scavenging activities) of developed products compared to the initial samples or non-augmented samples. Afterward, the extent of herbal incorporation, organoleptic properties, cooking quality, proximate composition and keeping quality of final products were also evaluated. Pertaining to the results, anti-glycation activity of instant rice was significantly increased (IC_{50} :37.6 μ g/ml); $p < 0.05$) after the herbal treatments. Anti-glycation activities of instant green gram (IC_{50} :146.7 μ g/ml) and

instant porridge mix (IC_{50} :33.4 μ g/ml) were also increased significantly compared to the herbal untreated samples. In addition, anti-oxidant activities of these developed products were also enhanced significantly ($p < 0.05$). The extent of herbal incorporated into the instant rice and instant green gram in terms of dry matter gain was 72.61% and 20.78%, respectively. The developed herbal treated products were organoleptically acceptable in accordance with the preferences provided by the respondents during sensory evaluation. The proximate compositions (carbohydrate, crude protein, crude fiber, fat, and ash) of developed products were also not changed significantly ($p > 0.05$) during the herbal treatment. There were no significant differences ($p > 0.05$) between the herbal treated and untreated instant rice as well as between the herbal treated and untreated instant green gram samples for the cooking quality parameters. Further, there was no significant difference ($p > 0.05$) between anti-glycation activities of developed products before and after the cooking according to the protocols. Under the keeping quality, herbal treated food products, packed with triple laminated packing material (PET/Al/LDPE) was chosen as the best packing material since it was not indicated remarkable increment in microbial growth (<100 CFU/g for both APC and YMC) as well as no significant differences ($p > 0.05$) in moisture increment, colour values (L^*a^*b) and anti-glycation activities compared to the initial levels, up to the 6 months of time period. Therefore, rice and green gram can be successfully augmented with anti-glycation activity and the developed instant rice, instant green gram and instant porridge mix products can be introduced into the market as anti-glycation activity augmented food products.