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Contents

NURSING STUDENTS' PERCEPTION REGARDING NURSING PROGRAM: A DESCRIPTIVE STUDY OF FOUR COHORTS

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ABSTRACT

Objectives: The aim of this study was to identify the nursing students' perception of nursing as a profession and factors influencing this perception. Methods: A descriptive cross-sectional design was used to achieve the study aim. A convenient sample of 120 nursing students enrolled in the nursing program at the college of Nursing, King Saud bin Abdul-Aziz University for Health Sciences, Riyadh participated in this study and classified equally to Pre-professional program years and professional program years. Self-Administered Questionnaire used for data collection. It consists of two parts: Part I include questions related to sociodemographic characteristics of the study subjects and Part II used to explore nursing students' perceptions of nursing as a profession with total scores ranging from 18 to 36, the greatest the score the more positive the perception. Results: The mean age of study subjects were 20.33+ 1.3 S.D. and the majority of them were single. The majority of students' families allowed them to join nursing college (72.5%). Around half of the participants were self-motivated to join nursing (40.8%). Regarding the perception of nursing as a profession there was a difference in mean score between the Pre-professional and professional program years (29.3 + 1.87 S.D. and 30.22 + 2.1 S.D. Respectively). The study results revealed a positive significance correlation between the level of education and perception ($r = 0.212^*$, $p = 0.02$). Conclusion: The nursing students' perceptions of nursing as a profession are positive among the majority of the study group and higher in professional program years than Pre-professional program years.

Index Term - Nursing Profession, Perception

I. INTRODUCTION

Nurses are health professionals who are the cornerstone of the health care structure. Efficient health services cannot be provided in health centers and hospitals without the nurse professionals. Unfortunately, there is a shortage of nurses worldwide above which posed a serious risk for patient safety, quality of care and public health as a whole. [1]. Nursing as a 'profession' is not just a definition, it is combination of a framework of communication and language of a group and this reflects who nurses

are and this meaning carries an individual's perception [2]. Nursing is defined as the optimal utilization of clinical decision to provide care to help people in improvement, maintenance, or recovery of health, in order to adjust with different health problems, and to attain the most excellent quality of life, especially in case of diseases and disabilities, and even death [3]. And also defined as the profession working to maintain, promote, and optimize health and abilities of individuals, prevention of diseases and harm, lessening of suffer through the proper diagnosis and accurate treatment of human problems, and engagement in the providing care and help for individual, family, community, and population [4]. Nursing is broadly regarding a dignified career in the community for the reason of the foundation significance in the life not only of the individuals but also of the community. Widely the nurse identified as a person who is the core stone of the health care system and the link between all the disciplines. Therefore nursing as a profession has the same importance to the different health care system [5].

Shortage of nursing has been a fact from the 70's to 90's century. This shortage was increased until reach a critical and difficult stage when compared to other jobs or carriers. [6]. Therefore there is a high need to increase the nursing recruitment in the different health care institutions and use of different strategy to retain them in the field of nursing. This is the main challenge nowadays in health care institutions worldwide [7]. Perceiving nursing as a profession and the choice to select nursing as a career for students is extremely affected by individual's past experience and ideas about the career. The community and media have a major role in the influence of the perception and thoughts the nursing student has regarding their career [8].

Students enrolled into the nursing colleges with intrinsic values of nursing that modify over time of education, allow them to be professional into the nursing career [9]. Several elements, namely educators, program, clinical teachers, nurses, friends, families, institutions and the environmental changes from the hospital to the different community settings form significant input to the progression of nursing students' professional acknowledgment [10].

The image of nursing as a profession is crucial in successful enrollment and retention of staff in the health-care organizations [11]. The perception of nursing students affecting their performance, image, resection to themselves, enrollment and retention in their nursing field. A very limited number of research studies addressed the perceptions of nursing students on nursing were found [12]. Student of nursing Who have perception about nursing and truths affecting which we fine searched in the Western region, lesser than investigated in the Arab Islamic countries [2]. In studies of nursing students 'perception of nursing as a profession, researchers investigated perception of students at different levels in an educational nursing program. Studies included assessment of student nurses' values and conceptions at entrance to nursing programs [13], after clinical practice [14] and before their graduation [15].

There are a huge number of literatures investigating the perceptions of nursing students all over the world, while there was a very little research from the gulf region. The literature emphasized that the students have a wide range of different entrance perceptions on nursing which have a high impact on their decision for continuation in nursing colleges [16].

In ancient time, nursing perception was negative. Ms. Florence Nightingale the, a well-educated woman from a wealthy class family became a nurse and improved the profession radically in nineteenth century that people gradually start to accept nursing as a respectable profession in the society [17]. For helping to

enrollments to the nursing carrier, a positive image of nurses and nursing has to be initiated though nursing education, nursing researches and the overall society [16].

II. SIGNIFICANCE OF THE STUDY

Kingdom of Saudi Arabia has been dependent mainly to expatriate nurses and this affecting the development of an indigenous nursing profession. Little information is known about Saudi student nurses 'perception regarding nursing as a profession and selected career. Clarification of the student's perception will help the educators and the authorized personnel for curriculum development to identify and evaluate the strength and weakness of their program as well as highlighting the factors that can affect this perception positively or negatively.

III. AIM OF THE STUDY

The aim of this study was to identify the nursing students' perception of nursing as a profession and factors influencing this perception

IV. RESEARCH QUESTIONS

1- How the nursing students' perceptive nursing as a profession?
2- Is there a change in nursing students' perceptions of nursing as a profession over the 4 years of the program?
3- What are the factors influencing 'perception of nursing as a profession?

V. MATERIALS AND METHODS

Research design: The current study used a descriptive cross-sectional design in order to answer the research questions.
Study Subjects: A convenient sample of 120 female nursing students enrolled in the nursing program at the college of Nursing, King Saud Bin Abdul-Aziz University for health sciences, Riyadh, from the female campus, participated in this study. The sample was selected equally in each cohort 30 students each, from the 4 years of the program, each cohort represented 2 levels. 1st cohort included level 1 &2 and 2nd cohort included level 3& 4; the 1st two cohorts represented students in pre-professional program. While the 3rd cohort included level 5 &6 and the 4th cohort included level 7& 8; the 2nd two cohorts represented students in professional nursing program. All willing students from the 4 cohorts and studying in the annual academic year 2014-2015 were eligible to participate in the study.
Data collection instruments: After reviewing related literature and to fulfil the aim of the study, one tool with two different parts was designed by the research team and revised by the consultants. The study tool was constructed, tested and piloted by the investigators to examine its applicability, clarity, reliability and feasibility for data collection, then revised by a panel of experts. Also content validity and expert's opinion were taken into consideration and the needed modifications were carried out and Face Validity of the tool was examined through a jury of three experts. The two parts are:
Part I
This part included questions related to socio-demographic characteristics of the study subjects such as sex, age, nationality, marital status, reasons for enrollment to the faculty of nursing, and family's opinion to students' enrollment.

Part II

It was used to explore nursing students' perceptions of nursing. It was scale format answered on yes or no responses: different nursing profession perceived concepts were used, the scoring of the positive perception was 2 if yes and 1 if no, while in negative perception, the 1 score for yes and 2 for the no response. The total scores of this questionnaire ranging from 18 to 38, the greatest the mark the more positive the perception. The cut off point for this scale was 27, if the total mark was 27 and above means positive perception, while if less than 27 indicated the negative perception.

Statistical analysis: Data were fed, coded, edited and analyzing using PC with statistical packages for social science (SPSS) version 22. Descriptive statistics were used numbers, percentage, arithmetic mean, standard deviation and correlation coefficient.

Procedure: Research proposal submitted to the research unit, college of Nursing, King Saud bin Abdul-Aziz University for Health Sciences, Riyadh and approval to start the current study were obtained. The data collected from the 4 equal cohorts (30 students each) at the same time. Official permission from the research unit obtained again to start the data collection, all the willing students were signed the consent, included all the ethical considerations for the current study, arrangement with teachers of each class were made to allow the researchers to take 15 minutes from the class for data collection. Each student spent around 10 minutes to fill her questionnaire. The researchers collected the data from the 4 cohorts at the same time and from each cohort until reaching 30 students.

VI. ETHICAL ISSUES

Informed consent was obtained from the students who agreed to participate in the study. The students' participation will be emphasized to be voluntary. Confidentiality and anonymity will be kept and students can withdraw from the study at any time without any affection of their grades.

Year 3 (Level 5 or 6)	**30**	**25.0**
Year 4 (Level 7 or 8)	**30**	**25.0**
Reason to join Nursing		
Secondary school GPA	**39**	**32.5**
Advice from others	**17**	**14.2**
Self-motivated	**49**	**40.8**
Forced by parents	**4**	**3.3**
Others	**11**	**9.2**
Family *monthly income*		
< 2500 SR	**5**	**4.2**
2500-5000 SR	**20**	**16.7**
> 5000-10000 SR	**43**	**35.8**
> 10000 SR	**52**	**43.3**
Family's opinion about the enrolment in the College of Nursing		
Agree	**87**	**72.5**
Disagree	**3**	**2.5**
Neutral	**30**	**25.0**

Habitation		
North Region	**17**	**14.2**
Central Region	**55**	**45.8**
Western Region	**12**	**10.0**
Eastern Region	**14**	**11.7**
South Region	**22**	**18.3**
Family member or relative in Nursing		
Yes	**53**	**44.2**
No	**67**	**55.8**

Table 2: perceptions on nursing as a profession

Nursing as a profession	Pre-professional program years (Year 1 &2) Number = 60		professional program years (Year 3 &4) Number = 60		Total Students Number = 120		Missed data
	Yes %	No%	Yes %	No%	Yes %	No%	
Helping others	100	0	100	0	100	0	-
An opportunity to serve the Humanity	93.3	6.7	100	0	96.7	3.3	-
An occupation not a profession	31.7	66.7	16.7	83.3	24.2	75	0.8
Enhancing personal growth	85	15	88.3	11.7	86.7	13.3	-
An appreciated profession in our society	45	55	58.3	41.7	51.7	48.3	-
A way to have a chance to travel abroad	53.3	46.7	66.7	33.3	60	40	-
A way to enjoy economic security	61.7	38.3	63.3	36.7	62.5	37.5	-
A dangerous and risky /dangerous profession	78.3	21.7	61.7	38.3	70	30	-
Caring profession with high ethical standards	90	10	96.7	3.3	93.3	6.7	-
An extremely hard profession	76.7	23.3	66.7	33.3	71.7	28.3	-

A respectful profession	88.3	11.7	63.3	36.7	75.8	24.2	-
Nurses just obey doctors' orders	45	55	20	80	32.5	67.5	-
Actually equal to other professions	53.3	46.7	50	50	51.7	48.3	-
An opportunity to get better marriage offers	50	50	25	75	37.5	62.5	-
Financially Rewarding profession	66.7	33.3	56.7	43.3	61.7	38.3	-
Helping health promotion and diseases prevention	90	10	91.7	8.3	90.8	9.2	-
Subordination to doctors	61.7	38.3	53.3	46.7	57.5	42.5	-
low academic standards	48.3	51.7	20	80	34.2	65.8	-
Total (Mean ± Standard Deviation)	29.3 ± 1.87		30.22 ± 2.1		29.76 ± 2.03		-

Table 3: Comparison between the different reasons to join Nursing and mean total score of perception

Reasons to join Nursing	Mean total score of perception
• Secondary school GPA	29.7179
• Advice from others	28.8824
• Self-motivated	30.1224
• Forced by parents	29.5000
• Others	29.7273
Total	**29.7583**

Table 4: Correlation Coefficient for age and Level of education in relation to the total score of perception among the all subjects (n=120).

Variables	r value	P - value
• Age	0.133	0.148
• Level of study	0.212*	0.02*

Demographics

Table 1 revealed that; The mean age of study subjects were 20.33+ 1.3 S.D. and the majority of them were single (91.7 %). regarding the Level of nursing study the students were equal in the 4 cohort (years) 25% each. Around half of the participants were self-motivated to join nursing (40.8%).The majority of students' families allowed them to join nursing college (72.5%). Family monthly income for around half of the students (43.3 %) was more than 10000 SR. as regards the family's opinion about the enrolment in the College of Nursing, the majority of them were agree (72.5 %). the highest percentage (45.8 %) of the students came from central region, Riyadh. more than half of the students didn't have Family member or relative in Nursing (55.8%).

Perceptions on nursing as a profession

Table 2 denoted that, the highest percentages of the students perceive the nursing as helping others (100%), an opportunity to serve the Humanity (96.7%), caring profession with high ethical standards (93.3%), helping health promotion and diseases prevention (90.8%) and enhancing personal growth (86.7%)

The mean score of perception was higher in professional program than pre-professional program years (30.22 $\pm$ SD = 2.1 & 29.3 $\pm$ SD = 1.87 respectively). Moreover the total mean score for all the students was (29.76 $\pm$ SD = 2.03).

Table 3 showed the comparison between the different reasons to join nursing and mean total score of perception and revealed the highest mean total score was between the self-motivated students as a reason to join the nursing (30.1224).

as shown in **Table 4**, the correlation coefficient for age and level of education in relation to the total score of perception revealed a positive correlation between the level of education and the perception ($r = 0.212^*$ at $p = 0.02$), while there was no correlation the age and the perception ($r = 0.133$ at $p = 0.148$).

DISCUSSION

The purpose of the current study was to identify the nursing students' changing perception regarding nursing profession over 4 years of the nursing program and generates an understanding of the factors influencing this perception at the College of Nursing, King Saud Bin Abdul-Aziz University for Health Sciences, Riyadh.

The study results indicated that, over the 4 years of the nursing program, nursing students generally have positive perceptions regarding the nursing profession, this finding is congruent with many studies in Egypt -2006 [18], Belgium-2010 [19], Bahrain, 2012 [16], [20] in Pennsylvania-1994 [21] and USA-1991 [22]. The current study denoted that, the highest percentage of the students perceive the nursing as helping others, an opportunity to serve the humanity, caring profession with high ethical standards, helping health promotion and diseases prevention and enhancing personal growth, this is supported by another study which showed that the majority of nursing students choose to study nursing because it opened opportunities for further professional development [23]. This is consistent with the study done by [24] and its results clarified that, from the student's point of view, nursing concerned caring behaviors and helping others and it was the most motive for the preference of nursing as a career as they mentioned.

In the same line a study done by [2] who added that, regarding the perception of the majority of nursing students towards nursing is a profession which offers opportunities to give care for people. These findings ensure that caring remains the chief spirit of nursing with a great value which cannot be ignored.

On the other side, the other researches were dissimilar to the current study results and revealed the following, [21] in Pennsylvania, mentioned that nursing carrier was attracting carrier because of job opportunities and availability. [20] in their study which done in Egypt and found that the majority of the students had selected nursing because of the good chance for job chance.

On the same line [16] in U.S.A reported that more than half of their study subjects enrolled to nursing colleges because of work availability. Matching with all of them [25] clarified that students almost choose nursing frequently because of the availability of job opportunities, financial security, proper salary, and motivation to join nursing.
As regard the students reasons to join nursing, the study results revealed that, around half of the participants were self-motivated to join nursing and in comparison between the different reasons to join nursing and mean total score of perception, the results denoted the highest mean total score was between the self-motivated students, in the same line [2] indicated that, It was expected to obtain students who select to study in nursing colleges as a primary choice and interest had more positive understanding of nursing profession than other .

This also is congruent with [26] in which their study findings revealed that, the decision for choosing nursing as a profession focus on favorite with awareness foresee of students who highly gratify with thought nursing with the other students who did not decide to be enrolled in nursing as a first choice for studying in the collage.

Regarding the total mean score for all the students towards the perception of nursing as a profession, it was positive with higher score in professional program than the pre-professional program years and there was a positive correlation between the level of education and the perception. Consistent with our results [27] mentioned which , Nursing students with past experience have the benefit of knowing imminent about the way of happening things in wards, justifying the fear of the unrecognized, and esteeming in the overall approach in comparison with students who have not any past experience of nursing.

In the current study, the majority of students' families allowed them to join nursing college and around half of the current study subjects have relatives or friends in nursing. this was matched with [28] who denoted that, the previous research showed that nursing students' images of nursing might be traced to relatives who work in health care and clarified that parents and friends were very powerful motivators for them to join nursing.

In congruent with the current study [29] they did investigation for the factors affecting the students selection of nursing as their career and clarified that the presence of any of the student's family members, friends or relatives in nursing field had a major impact on the choice and motivation to enrol in nursing colleges and to select nursing as a career.

CONCLUSION

The study concluded that, the nursing students generally have positive perceptions of nursing as a profession and it was higher in professional program years than Pre-professional program years. It means the perception is changing towards positive side. The study also concluded that. The majority of students' families allowed them to join nursing and around only half of the participants were self-

motivated to join nursing

RECOMMENDATIONS

The current study recommends conducting further studies on the perception of the nursing students in other different regions in Kingdom of Saudi Arabia, and to include the internship students. The factors that can affect the student's perception have to be considered during registration and admission.

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EXPERIMENTAL DESIGN ANALYSIS OF ELECTRIC POWER GENERATION AND REFRIGERATIONBY USING WASTE HEAT

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A B S T R A C T

Electricity is most important for the human life and industries, but the available energy is very less. Today, the demand of energy is increasing tremendously, but available energy lacks in supply. We are proposing a solution through this paper. The main stress of this paper is on energy conservation by using technique of utilizing waste heat from exhaust waste heat from IC engines (automobiles). This power can be used to charge the battery and reduce the fuel consumption and battery life is also increased. The paper includes the design and analysis of TEG with the material used; efficiency of TEG and the heat transfer in TEG are also studied using ANSYS. The study shows the factors affecting the efficiency of TEG. The main objective of this paper is to produce power from the waste sources and to reduce the heat let into the atmosphere, reduce the fuel consumption and to increase the life of the battery used in automobiles.

Index terms - Energy, TEG, Efficiency, IC engines

I. INTRODUCTION

Energy is a basic requirement for the existence and development of human life. Waste heat recovery and utilization is the process of capturing and reusing waste heat for a useful purpose. Waste heat which is rejected from a process at a temperature enough high above the ambient temperature permits the recovery of energy [1].This invention relates to the Internal Combustion Engine. Among all research directions, waste heat recovery (WHR) is most concerned, due to the widespread existence and high accessibility of suitable resources. According to India Bureau of Energy Efficiency, the benefits of WHR includes reduction in the process consumption and costs, reduction in pollution and equipment sizes, and also reduction in auxiliary energy consumption[2] . From the research found out that by amount of electrical energy is found from domestic refrigerator and the hot case can be saved. We found recovery of heat from the condenser reduces the heat load to surrounding and makes surrounding comfortable from the research [3]. By using the method of energy conservation can improves the thermal efficiency of the system should be obtained [4].From the research paper [5], the pyroelectric waste heat should be produced from the waste heat conduction. In this paper authors have investigated and proved the electricity produced from refrigeration waste heat with the aid of thermoelectric generator. The thermo electric generator (TEG) is the system used to generate power from low temperatures that is less than 1000K as heat input. It is best method for recovery of exhaust heat. It can be used in many fields such as automobiles, boilers wood stoves etc. The efficiency of a TEG depends on the thermo electric materials.
Waste heat is heat, which is generated in a process by way of fuel combustion or chemical reaction, and

then “dumped” into the environment even though it could still be reused for some useful and economic purpose. This heat depends in part on the temperature of the waste heat gases and mass flow rate of exhaust gas. Waste heat losses arise both from equipment inefficiencies and from thermodynamic limitations on equipment and processes. For example, consider internal combustion engine approximately 30 to 40% is converted into useful mechanical work. The remaining heat is expelled to the environment through exhaust gases and engine cooling systems [7]. A thermoelectric generator (TEG), also called a Seebeck generator, is a solid state device that converts heat flux (temperature differences) directly into electrical energy through a phenomenon called the Seebeck effect. Thermoelectric generators function like heat engines, but are less bulky and have no moving parts. A battery is employed to store the electrical power generated by the thermoelectric generator. The electric power from the battery is used as the input of the thermoelectric cooler. The working of the refrigeration system based on peltier effect. The device called thermoelectric cooler. In this refrigeration system there is no compressor, gas is not being used. The cooling is obtained from the cooler through the following procedures. The project describes the design and analysis of TEG and TEC with the material used, properties, factors affecting efficiency is analysed both experimentally and through the “ANSYS” software.

II. MATERIALS AND METHODS

Waste heat is heat, which is generated in combustion process or chemical reaction, and then flow into the environment even though it could still be reused for some useful and economic purpose. The components used to reuse the waste heat are Compressor, Condenser, Expansion valve, Evaporator, Thermoelectric Generator, and Battery. Mainly Compressor needs work, W. The work is supplied to the system. Condenser is used to reduce the temperature of a system and emits the heat. There is no exchange of heat during throttling process through the expansion valve as this process occurs at constant enthalpy. Thermoelectric generator is governed by simple solid state devices called thermoelectric power generators which are capable of conversion of heat energy into electrical energy. Thermoelectric generators convert heat into electricity through the Seebeck effect. However, thermoelectric generators have lower efficiency when compared to many other power generation devices. The generated electrical energy stored by the battery. When a temperature difference is applied between the two junctions of two dissimilar metals, a voltage is generated which is called the Seebeck Voltage and this phenomenon is the guiding principle behind thermoelectric power generation. Two dissimilar metals maintained at different temperature, when connected by a wire, will produce potential difference.

BLOCK DIAGRAM

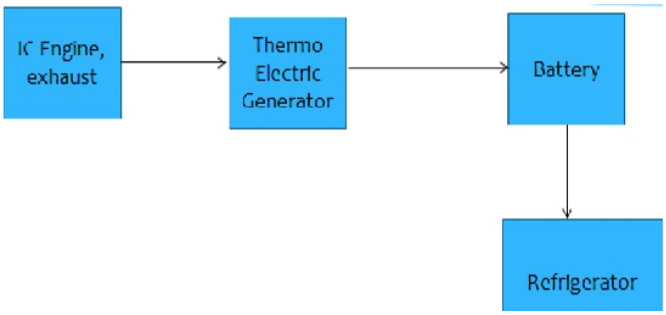


Fig.1 Block Diagram of Experimental setup

III. EXPERIMENTAL SETUP

The experimental setup for testing is shown in figure

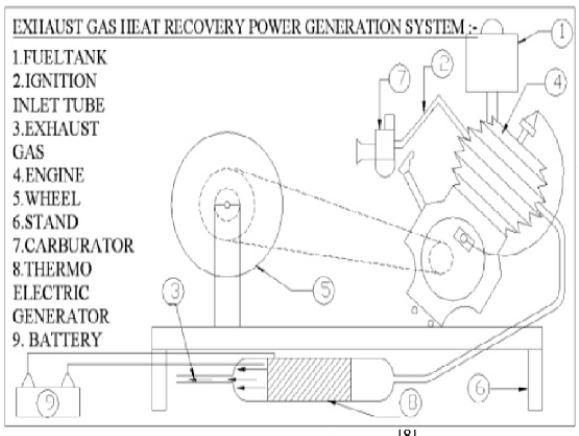


Fig.2 Experimental setup [8]

DESIGN OF EXHAUST WITH THERMOELECTRIC GENERATOR

The following is the design of the exhaust done in the Ansys workbench software. Given design shows the schematic diagram of thermoelectric generator header portion clamped on exhaust cylinder of 37.5mm inner diameter and thickness is 1mm.

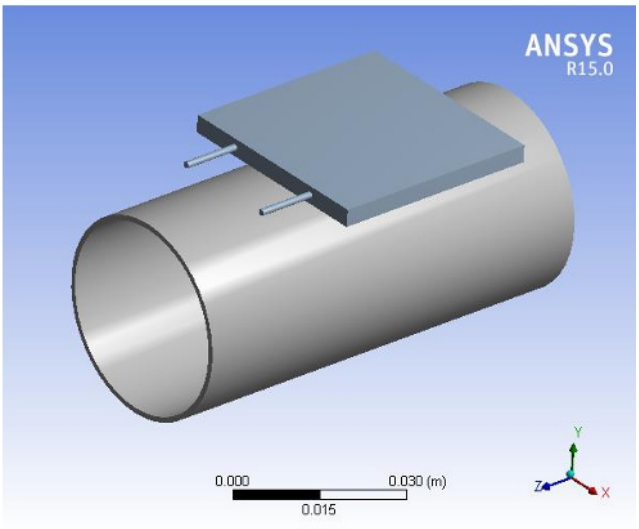


Fig.3 schematic diagram of thermoelectric generator

DESIGN OF THERMOELECTRIC REFRIGERATOR

The thermoelectric refrigerator is used for Cooling of system electronically by using the peltier effect i.e. heat is pumped with electrical energy using thermoelectric modules.The design of thermoelectric refrigerator with the chamber drawn in Ansys workbench with dimension 122*80*30 mm.

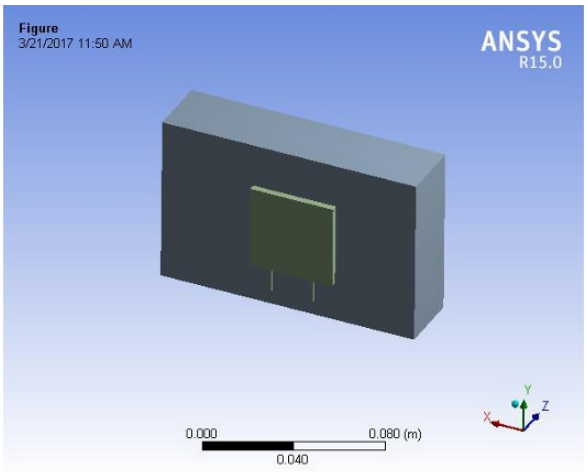


Fig.4 schematic diagram of thermoelectric refrigerator

DESIGN CALCULATIONS

The material is selected on the basis of power factor & melting point. The performance of the thermoelectric material can be expressed as

$$Z=\alpha 2/kR$$

Where, ΔV is change in voltage. ΔT is change in temperature.
R is the electric resistivity and k is the total thermal conductivity The thermal electrical resistivity of bismuth tellurium is = 1.29μΩm
Where, ΔV = max. volt.- min. volt.

12 – 1.36 =10.64v
ΔT = max. Temp. – Min. temp.
140 - 40 = 100
α=-{ΔV/ΔT}, -(10.64/100) = -0.1064v/k
α= - 0.1064v/k , α2 = 0.01132 v/k

The total thermal conductivity k=7.97 w/mk
Therefore, Z = (0.01132 /(7.97*1.29))= 1.101*10-3
Thus the performance of thermoelectric material is,
Z = 1.101*10-3

IV. RESULTS AND DISCUSSIONS

TEMPERATURE OF GENERATOR

The following design specifies the temperature variation in the generator and the maximum voltage obtained from the single module is 4volt,as per increase in number of module the power obtained will be also high.

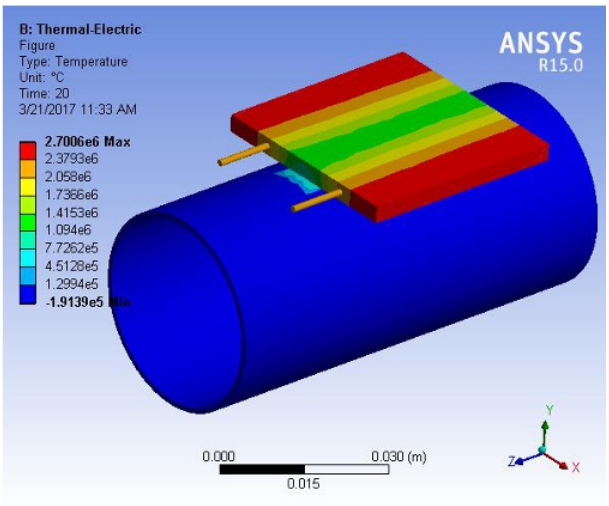


Fig.5 shows the temperature variation in the generator

ELECTRIC VOLTAGE

The electric voltage which is obtained by three modules is shown in the design, 4volt is the maximum obtained from single module, as per increase in the number of module the power generated will be high.

ΔT = max. Temp. – Min. temp.

140 - 40 = 100

$\alpha = -\{\Delta V/\Delta T\}$, -(10.64/100) = -0.1064v/k

α= - 0.1064v/k , $\alpha 2$ = 0.01132 v/k

The total thermal conductivity k=7.97 w/mk

Therefore, Z = (0.01132 /(7.97*1.29))= 1.101*10-3

Thus the performance of thermoelectric material is,

Z = 1.101*10-3

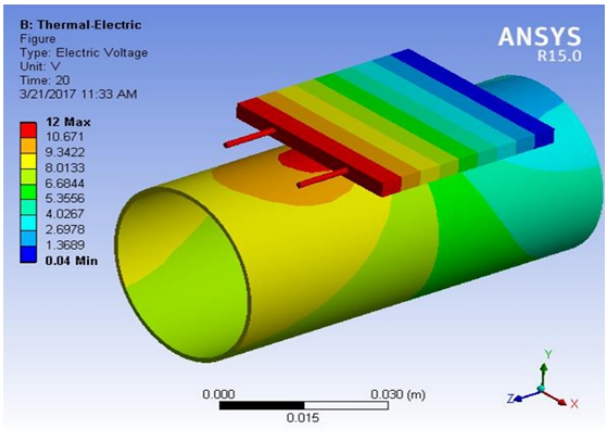


Fig.6 shows the voltage variation in the generator

TOTAL CURRENT DENSITY

The schematic design defines that the total current density which has been Obtained at the time of 20 minutes at the maximum and minimum range of span

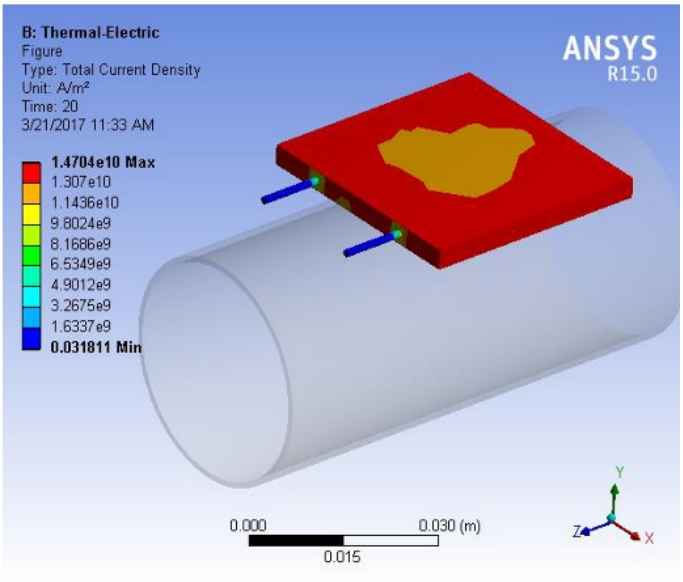


Fig.7 shows the total current density of generator

TRANSIENT TEMPERATURE
THERMOELECTRIC REFRIGERATOR

The term transient temperature means the flow of temperature in between the two solid materials. In the following representation there is the maximum and the minimum occurrence of temperature difference from the following time span of 20 minutes 18ºc is the maximum chillness is been obtained from thermoelectric refrigeration system.

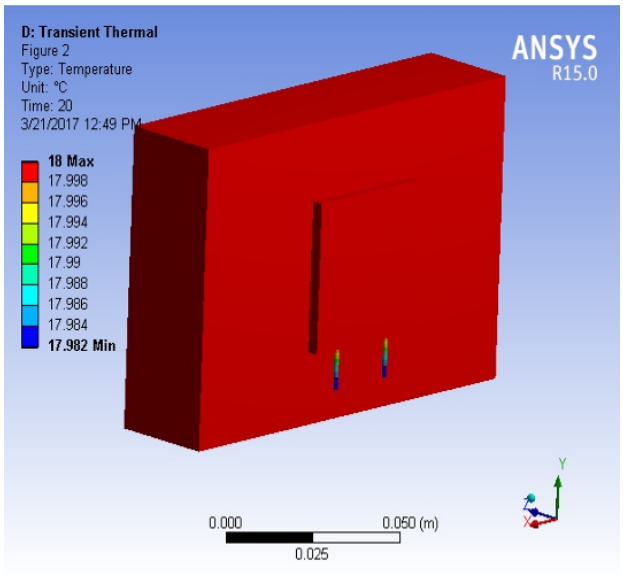
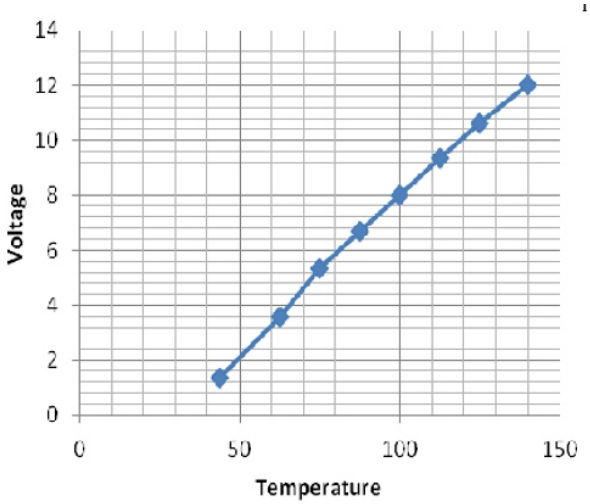


Fig.8 shows the transient temperature of generator

The graph shows the output reading of voltage with the variation in temperature, if temperature is increase voltage also increased and able to obtained 12volt at 140.The data obtained analytically through ANSYS Thermoelectric& Transient thermal software.



Graph.1 shows the output voltage with temperature variation

CONCLUSION

Thus the objective, generation of electric energy from waste heat from IC engine for the purpose of refrigeration system has been analyzed analytically, also calculated to generate electricity from thermal energy as alternative sources. Thermoelectric power generators make it capable of largely reducing capital cost, increasing stability, saving energy source. We conclude that TEG system of charging the battery could reduce the fuel consumption and also battery life used in automobiles could be increased. It is the best method for recovery of waste exhaust heat.

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DEVELOPMENT AND VALIDATION OF ANALYTICAL METHOD FOR ASSAY DETERMINATION OF ISOSULFAN BLUE BY LIQUID CHROMATOGRAPHY

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A B S T R A C T

A novel, simple and rapid method has been developed and validated for quantitative assay determination of isosulfan blue in drug substance by Ultra Performance Liquid Chromatography (UPLC). Analytical method was developed and validated as per ICH guidelines. Reverse phase liquid chromatography method is developed and validate on UPLC using C18 (100 x 2.00 mm) 1.9µ column. 0.1 % perchloric acid in mixture of 73 volumes of water and 27 volumes of acetonitrile as mobile phase. The ultra violet wavelength selected 220 nm, the column temperature 30ºC. Linearity study was carried out for isosulfan blue linearity was calculated from 80% level to 120% with respect to specification level. The correlation coefficient r = 0.999 was proved and showed that the method is linear. Specificity was studied by individual injection of drug substance, related impurities and spiking study as well. It was evident from specificity test that all the related impurities are separated from isosulfan blue peak and method is specific. Isosulfan blue sample solution and mobile phase were found to be stable for at least 3 days.

Keywords - Isosulfan blue, UPLC, Assay, Validation, Reverse phase, Solution stability.

I. INTRODUCTION

Isosulfan blue is used as a cancer diagnostic agent [1].It is having chemical name, N-[4-[[4(diethylamino) phenyl] (2,5-sulfophenyl) methylene]2,5-cyclohexadien-1-ylidene]-N-ethylethanaminium, sodium salt [2]. Other names are sulfan blue or patent blue is an active pharmaceutical ingredient (API's) [2].Chemical formula is C27H31N2NaO6S2, chemical structures show in **Fig.1.**

In the literature, there is no analytical method was reported for assay determination of isosulfan blue. This report describes a reverse-phase ultra performance liquid chromatography method for the assay test for isosulfan blue on by using C18 (100 x 2.00 mm) 1.9µ column. The developed UPLC method was validated for assay determination of isosulfan blue in drug substance as per validation of analytical procedure guidelines [5].

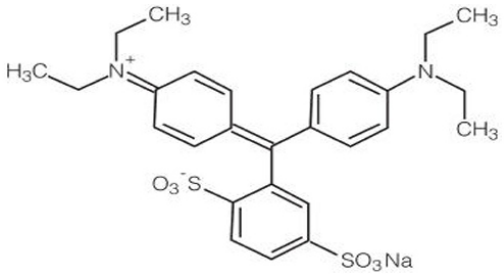

Fig.1: Chemical structure for isosulfan

II. DETAILS EXPERIMENTAL

2.1 Materials and Procedures

Following materials are used to develop and to validate the analytical method. The chromatographic parameters are fixed after development of the method and are validate.

2.1.1 Chemicals

Isosulfan blue sample, standard and impurity-A, impurity-B and impurity-C were kindly gift by Emcure Pharmaceuticals Ltd Pune, Maharashtra, India. Perchloric acid (AR grade) was purchased from JT baker, HPLC grade acetonitrile was purchased from biosol, sodium dihydrogen phosphate dihydrate (AR grade) was purchased from merck and millipore water is used for mobile phase preparation and Diluent preparation.

2.1.2 Equipment

Waters UPLC having make acquity system inbuilt with auto injector was used for method development and validation. Empower software was used for data acquisition and system suitability calculations.

2.1.3 Standard preparation

Weigh accurately about 50 mg of isosulfan blue standard in 50 ml volumetric flask dissolve in diluents and dilute up to the mark (stock solution), further pipette out 5 ml of stock solution and dilute 50 ml volumetric flask.

2.1.4 Sample Preparation

Weigh accurately about 50 mg of isosulfan blue sample in 50 ml volumetric flask dissolve in diluents and dilute up to the mark (stock solution), further pipette out 5 ml of stock solution and dilute 50 ml volumetric flask.

2.1.5 Chromatographic Conditions:

The chromatographic conditions were optimized by using column C18 (100 x 2.00 mm) 1.9µ column. The flow was set as 0.35 ml/min. The column oven temperature was maintained at 30ºC and the detection was carried out at the wavelength of 220 nm. The injection volume is 2µl.

2.1.6 Mobile phase

0.1% perchloric acid in mixture of 73 volumes of water and 27 volumes of acetonitrile.

III. RESULTS AND DISCUSSION

The developed assay method of isosulfan blue has been validated as per ICH guideline [5]. All the parameters are covered during validation and found within acceptance limit.

3.1 Specificity

Specificity study was carried out to verify there is no interference with analyte (isosulfan blue and all known impurities) peak is observed from the blank solution. All the impurities peaks are well separated

from isosulfan blue and peak purity of isosulfan blue peak was found complying. Typical chromatogram is showed in **Fig.2.** The results are tabulated in Table

Table 1: Results of Specificity of isosulfan blue

Injection Number	Peak area of isosulfan blue
1	1255540
2	1255466
3	1264262
4	1252779
5	1262085
6	1256177
Mean	1257718
(%) RSD	**0.4**

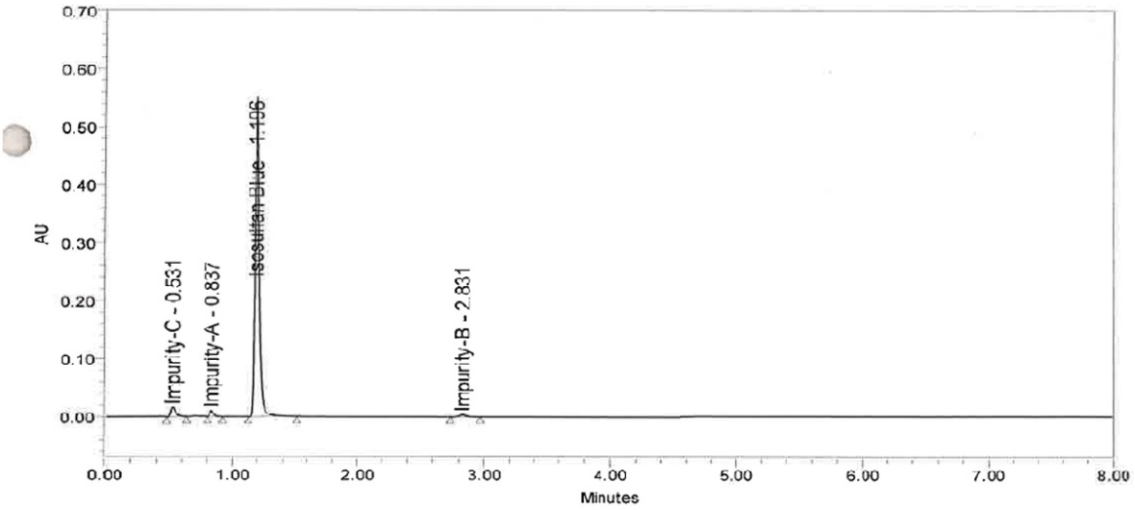

Fig.2: Typical UPLC chromatogram with data of system suitability (spiked sample)

3.2 Forced degradation

As part of specificity study, product was subjected for degradation under different conditions like basic, acidic, oxidation, thermal, photolytic, humidity conditions. In degradation study, degradants found well separated from the peak of isosulfan blue. It is also found that all impurities are separated from isosulfan blue.

Table 2: Forced degradation study results

Condition	Degradation achieved in %	Peak purity angle for isosulfan blue	Purity threshold for isosulfan blue
Acid degradation	19.758	0.087	0.293
Base degradation	22.865	0.080	0.288
Peroxide degradation	11.089	0.081	0.279
Thermal degradation	1.047	0.080	0.293
Photolytic degradation	0.971	0.081	0.289
Humidity degradation	1.050	0.077	0.288
Untreated (as is)	-	0.096	0.290

3.3 Linearity and range

Linearity was calculated from 80% to 120 % with respect to specification level for isosulfan blue. The range was selected from 80%, 90%, 100%, 110% and 120% of specification limit for isosulfan blue.

Table 3: Linearity of isosulfan blue results

Linearity level	Concentration (in ppm)	Mean area
Level 1 (80%)	74.238	969251
Level 2 (90%)	83.518	1095766
Level 3 (100%)	92.798	1224317
Level 4 (110%)	102.078	1349977
Level 5 (120%)	111.358	1469069
correlation coefficient (r)		**0.999**

3.4 Precision System precision

Blank solutions and six replicate injections of standard solution were injected as per methodology. % RSD of area obtained from six replicate injections of standard solution was calculated. Results are tabulated in Table 4.

Table 4: System precision results

Injection Number	Peak area for isosulfan blue standard
1	1219866
2	1215330
3	1216390
4	1218471
5	1222431
6	1219296
Mean	1218631
SD	2544
% RSD	**0.2**

3.5 Method precision

Method precision was demonstrated by assay test of six replicate samples at specification level. Calculate relative standard deviation for isosulfan blue. Results are tabulated in **Table 5.**

Table 5: Method precision results

Injection Number	Area of isosulfan blue	%Assay (n=6)
1	1242727	99.624
2	1244653	99.902
3	1251494	99.851
4	1247154	99.899
5	1243390	99.946
6	--	99.935
Mean	1245884	99.860
SD	3563	0.1203
% RSD	**0.3**	**0.12**

3.6 Intermediate precision

Intermediate precision of the analytical method was determined by using method precision parameters on different days, by different analysts, on different instrument and by different column lot number using the same sample used in method precision. Analysis was performed as per methodology, under same experimental condition by injecting six replicate sample preparations. %RSD of results was calculated. Overall % RSD of method precision and intermediate precision results was also calculated. The tabulated data shows that the Analyst–I have done the method precision and Analyst –II has done the intermediate precision. Results are tabulated in Table 5, Table 6 and Table 7.

Table 6: Intermediate precision results

Injection Number	Area of isosulfan blue standard	%Assay (n=6)
1	1213296	99.778
2	1225007	99.726
3	1223621	99.797
4	1216396	99.737
5	1217692	99.782
6	---	99.823
Mean	1219202	99.774
SD	4956	0.0364
% RSD	**0.4**	**0.04**

Table 7: Overall % RSD for method and intermediate precision

Overall RSD of method precision and intermediate precision results (n=12)		
Compound	**Mean (%)**	**Overall RSD (%)**
isosulfan blue	99.817	0.10

3.7 Solution stability

Solution stability was performed at the room temperature. Test sample was prepared at specification level and injected as fresh, 1 day, 2day and 3day intervals.

Table 8: Solution Stability results

Sr. No.	Interval	% Assay	Cumulative % RSD
1	Sample Preparation Fresh	99.492	-
2	Sample Preparation, 1 Day Old	99.507	0.01%
3	Sample Preparation, 2 Days Old	99.579	0.05%
4	Sample Preparation, 3 Days Old	99.606	0.06%

3.8 Robustness

Robustness of the method was verified by varying the instrumental conditions such as by changing flow rate ± 10% (flow= 0.385 mL/min & 0.315 mL/min) by changing column oven temperature ± 5°C (temp.= 35°C and 25°C) by mobile phase concentration ± 2% (buffer 71+29 acetonitrile and buffer 75+25 acetonitrile) and by changing column lot number. The system suitability was evaluated in each condition and sample was analyzed in triplicate.

Table 9: Robustness Study results

Robustness study	% RSD	Mean %Assay
Column lot-I	0.24	99.798
Column lot-II	0.06	100.055
Flow+10%	0.03	99.942
Flow-10%	0.04	99.843
Mobile phase composition +2%Acetonitrile	0.10	100.001
Mobile phase composition - 2%Acetonitrile	0.12	99.388
Column oven temp +5°C	0.08	99.577
Column oven temp -5°C	0.58	99.918

CONCLUSION

Developed and validated rapid Assay method for isosulfan blue drug substances by UPLC method as per ICH guidelines.UPLC is modern technology for chromatographic analysis for fast analysis. All the impurities and principal peaks are eluted within 5 minutes. All the validation parameters found well within acceptance criteria. The developed method is suitable pharmaceutical industries quality control laboratory and research laboratory.

ACKNOWLEDGMENTS

Above mentioned research work required many requisites like standards, samples, impurity standards and analytical support. All this support was provided by Emcure Pharmaceuticals limited, Pune, Maharastra.

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MEMBRANE PROCESSES APPLICATIONS FOR TREATMENT OF YEAST INDUSTRY WASTEWATER

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A B S T R A C T

Bread yeast process wastewaters have dark brown wastes from melanin which used as the main raw material. They contain of high organic pollutant concentrations such as high COD, dark color, nitrogen and refractory organic components. These components can be removed by conventional treatment methods. For that reason, in recently years new treatment technologies have been developed and applicate. Thanks to those systems, these wastewaters could be both treatment and reuse for particularly irrigations of agriculture areas. so, stringent legal limitations to be requested for treating of wastewaters could be provided as well as using irrigation of agriculture areas

Keywords- Biological treatment, yeast wastewater, membrane bioreactor, reuse, irrigation of agriculture areas.

I. INTRODUCTION

Yeast industry produce wastewaters highly rate during production of the yeast. These wastewaters consist from operation process, washing of production areas and reactors. The wastewaters could not be discharge directly for dense pollution. They need to treatment. Without treating, they don't provide discharge standards. Initially, both aerobic and anaerobic treatment methods have been used. However, because of strict discharge standards, those methods stay inadequate. In last years, membrane processes have been consisted to be added membrane to those systems.

Biomass accumulation can be provided highly by membrane processes and can be operated at higher concentrations (S. Maredia, 2015). Also, they have provided highly quality effluent for wastewater treatment of that and so on (Aslan and Saatçı, 2013). in case of operation under anaerocik conditions especially, their advantages can increase (B.Q. Liao et al., 2006; Aqunio et al., 2006). in such case, treatment can be provided as well as bioenergy production from domestic and industrial waste and wastewaters (P.Le-Clech, 2010).

In regionals of being fresh water scarcity, treated water can be used to irrigation of agriculture areas; even it can also be used as drinking water. So, water the used by for drinking water is saved. as known, the largest amount of fresh water has been used for agriculture and landscape irrigation (FAO ,2003; A. Alobaidy et al., 2010). Applications of treated wastewater for irrigation of agriculture and landscape areas have benefits for both environmental and economic impacts. For example, water shortage could be

prevented, especially, in arid regionals, it could provide drinkable water from high quality sources, Nitrogen and phosphor nutrients within wastewater can be increased efficiency on agricultural applications (C. Lubello et al., 2004). Also, the treated waters could be implicated in a lot of different areas such as toilet washing, park garden water, reuse in the system, underground feeding and so on.

II. YEAST INDUSTRY WASTEWATER CHARACTERISTICS

Yeast industry wastewaters are of acidic feature and high organic pollution and COD values. Also, theirs color is quite dark and intense. High molecular weight components usually cause colored compounds. These components are source of molasses. These components are usually caused by molasses production, which is used as raw material in bakery yeast production. The main pollutant parameters in the yeast industry wastewater are COD, BOD, BOI, color, bad smell, oil-grease, total nitrogen and phosphorus. Phenol and sulphates and pathogenic microorganisms may also be present (Tünay, 1996). Yeast industry wastes contain various pollutants. The characteristics of the effluent for a plant with a twostage treatment system are given in Table 2. Although the color value is not measured for operation, the outlet water is always very dark.

Table 2. The characteristics of the effluent for a plant with a two-stage treatment system as Aerobic-Anaerobic (Pala et al., 2005)

Parameter	Process outlet waters (Anaerobic input)	Anaerobic output (Aerobic Input)	Aerobic output (Discharge)
pH	5.9-6.12	7.6-8	7.6-8
COD (g/L)	20.1-22.1	3.8-4.4	0.6-0.9
BOI5 (g/L)	14.1-15.4	2.3-3.0	0.09-0.15
Oil grease (g/L)	0.009-0.01	0.009-0.01	0.009-0.01
Color	High dark	Dark	Clear

There are generally three types of wastewater in the yeast industry. The first is wastewater with a high pollution concentration of 8-10 g /L of COD. This wastewater occurs in the yeast production system, especially in the purification, centrifugation and filtration of the yeast. Secondly, KOI values are moderately polluted wastewaters in the range of 0.5-3 g / L. They consist of process and demolition waters. The third is wastewater with a KOI value of less than 0.5 g / L. Various washing operations result from operations such as equipment rinsing, boiler blowing. The colors are clear.

III. MEMBRANE APPLICATIONS IN MAYA WASTEWATER TREATMENT

Biological treatment methods are used because yeast industry wastewater, which is one of the important food industries, contains a wide variety of pollutants characterized by high chemical oxygen demand (COD), high biochemical oxygen demand (BOD), dark color and biodegradable organic compounds. As is known, biological treatment methods are the most common methods for removing organic compounds in wastewater. However, these biological treatment methods may not be sufficient to remove biological hardly degradable organics and coloring in the wastewater. Therefore, the development of the biological methods or different methods is investigated to provide discharge criteria for the removal of biologically hard degradable organics in wastewaters. Generally, two stage treatment as anaerobic-aerobic units are applicate for the yeast industry wastewater. However, even after this application, while the COD removal in process water is provided to a large extent, the dark color

originates from molasses, which is the raw material of yeast, production cannot be removed. In cases where sufficient color removal is not achieved, yeast wastewaters given into the receiving water environments, in which both impairs the aesthetic appearance and reduces the light permeability. Reducing the light permeability causes both decreasing of the dissolved oxygen amount and suffering of living things. Also it restricts water reuse possibilities.

Mutlu et al. (2001) studied the membrane processes with the aim of providing color removal from the wastewater of the yeast industry. MF, UF and NF membranes were investigated in this study. The study was used wastewater with a COD value of 4g / L. The wastewater with 11000 Pt-CO color and 2.4 optical density value was first applied to the MF process followed by NF. In the study, COD removal was 72%, color removal was 89% and optical density recovery was 94%. They said that the resulting filtrate was being completely clear.

In a study conducted by Pak (2011), for treatment of wastewaters of Pakmaya bakery production plant in Marmara Region in Turkey membrane processes such as MF, UF, NF and RO was investigated. this researches were applicate for effluent waters of two stage treatment (Anaerobic +Aerobic).Thus, an advanced treatment was tried. Four stages have been applied in the experimental study. These steps are given in figure 1.

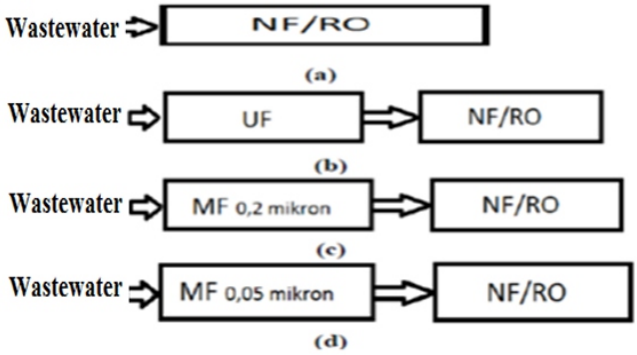

Figure. 1. Treatment steps that can be applicate for the treatability of wastewaters of baker's Yeast Plant

NF and RO processes were operated at 12-15 bar and at 5-30 bar pressure, respectively. Color, COD, pH, temperature, salinity, conductivity, total solute removal and flux recovery of the instant samples taken from filtration and concentration stream were investigated in the studies. Also, flux and transmembrane pressure were observed.

In UF + NF application under 15 bar pressure, 75% COD, 96% BOI5, 93% AKM, 14% conductivity and 20% salt removal have been provided. In the work done with the NF process, over 70% COD removal have been achieved. When this process is used with UF or MF, the COD removal has reached 95-99%. Color removal was 99%. In the RO processes, it is stated that the formation of concentrate flow is high. MF/UF+ NF application for treatment of yeast industry wastewater has been reported to yield optimum efficiency.

In a study conducted by Yalçın (1998), color, COD and ammonia nitrogen removal from yeast industry wastewaters have been studied by RO membrane process. The wastewater with COD of 2.4-3.3 g / L, ammonia content of 0.4-0.7 g/L, conductivity of 6310-10100 µS/cm and color value of 4500-5200 PtCo

was obtained in the study. Here have been provided 98-99% COD removal, 93% ammonia removal and 97-98% color removal. In a study by Saeki et al., (1992), treatability by membrane filtration of wastewaters of bread yeast which is used to molasses as raw materials was investigated. Membrane filtration was applied to depending on the properties of pollutant components with different molecular weights. When the wastewater was applied to the membrane process with a range of 200.000-300.000 Da, the color removal is determined as 74%. While wastewater applied to the 40.000 Da membrane process, 56% color removal is provided. When wastewater was applied to a membrane process with 20,000 Da, the color removal was 78%. Finally, when wastewater

was applied to the 10,000 Da membrane process, the color removal increased ın up to 85%. Thus, it has been suggested that higher yields will be achieved if the permeability of the applicable membranes in the color removal is less than 30,000 Daltons.

Membrane studies were carried out using a pilot scale membrane filtration system. this system was operated with a spiral wound membrane module with a membrane surface area of 2 m2 at a pressure range of 1-70 bar. The temperature of the system was set to 25-270C. UF and RO membrane processes have been applied to treat wastewater discharge. Since the pilot membrane system is operated in closed mode for both UF and RO applications, concentrate and filtrate flux were returned to the feed tank. The wastewater was pumped from a 60 liter feed tank to a spiral wrapped module (Figure 2)

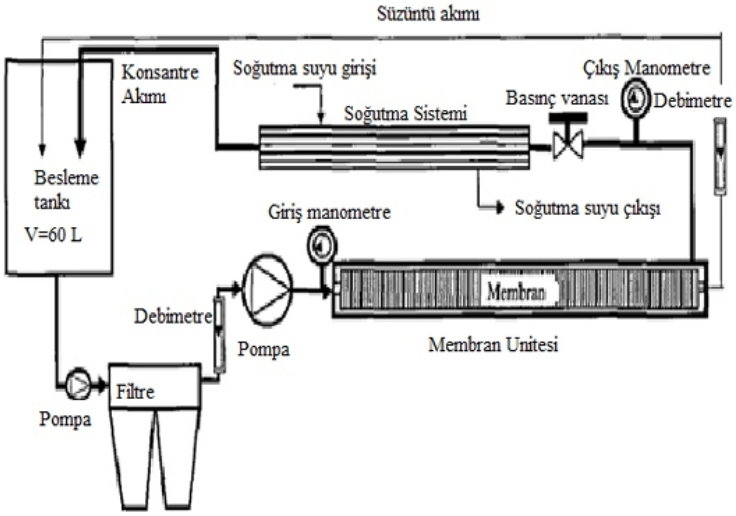

Figure 2. Flow diagram of Pilot scale membrane system.

As a result of this pilot application, the following data were obtained.
*It has been found that UF and / or RO membrane treatment systems for yeast wastewater with high pollution are efficient in COD, color, nitrogen and conductivity removal.
• The ultimate disposal of the concentrated part of originating from the membrane system has been an important problem. Therefore, there is a need to additional advanced applications. The biodegradability of the concentrated part formed in the RO system can be increased significantly by applying advanced oxidation of ozone and / or hydrogen peroxide. Additional COD removal can be achieved in the existing biological treatment plants with the oxidized concentrate fraction.

CONCLUSIONS

Thanks to membrane processes, yeast industry wastewaters could be both treatment and reuse for particularly irrigations of agriculture areas and other so areas. Therefore, stringent legal limitations to be requested for treating of wastewaters could be provided and reuse different areas.

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GONADAL AND EXTRA-GONADAL SPERM CHARACTERISTICS OF RABBIT BUCKS FED WITH DIETS CONTAINING RAW OR FERMENTED COTTON SEED CAKE SUPPLEMENTED WITH GINGER (ZINGIBER OFFICINALE ROSCOE)

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A B S T R A C T

The potential of cottonseed cake (CSC) as a veritable source of protein, energy and fibre for farm animals has been demonstrated. However, its use is limited to ruminant feeding due to the presence of gossypol, a polyphenolic compound of significant physiological implications. Fermentation is one of the biotechnological methods for detoxifying a variety of feed ingredients. This study investigated the effect of raw or fermented CSC – based diets with or without ginger supplementation on gonadal and extra-gonadal sperm characteristics of rabbit bucks. Thirty (30) cross-bred (New Zealand White X Chinchilla) bucks, 6 -7 weeks old averagely weighing 768.54g, were randomly assigned to five dietary treatments (n = 6 per treatment) in a 2 X 2 factorial within a completely randomized design (CRD). The CSC replaced soyabean meal (SBM) at 0% (control) and 100% (Raw or Fermented) with or without ginger supplementation (30mg/kg feed). Animals were fed for 12 weeks and slaughtered. Their reproductive organs were processed for sperm evaluation. Raw CSC without ginger supplementation resulted in lower ($p<0.05$) sperm concentration, Gonadal Sperm Reserve (GSR) and Daily Sperm Production (DSP); and higher ($p<0.05$) dead sperm than other treatments. The sperm concentration (69.33 X 106/ml), motile sperm (79.97%), GSR (66.44 X 106) and extra-gonadal sperm reserve (109.98 X 106) were highest for bucks that consumed fermented CSC with ginger supplementation. This study shows that total replacement of SBM with raw CSC reduced sperm quality in rabbit bucks. This adverse effect was corrected by a combination of fermentation and ginger supplementation.

Keywords- Rabbit, Cottonseed cake, Fermentation, Ginger, Sperm characteristics

I. INTRODUCTION

Cottonseed cake (CSC) is a cheap and excellent source of animal protein, energy and fibre for variety of animal species [1]. The CSC is one of the feedstuffs that have not been fully exploited for livestock feeding, most especially non-ruminant animals despite its high nutritional potential. Its crude protein content ranges from 35 to 46%, fibre 11-13% and TDN 70-80% [2]. The high nutritional profile of CSC notwithstanding, it is only used for ruminant feeding. Non ruminant animals are not usually fed with CSC because of the presence of gossypol, a toxic factor which has been known for anti-fertility effects on male animals. Extensive research has demonstrated a variety of toxicological effects of gossypol on

male reproductive characteristics of farm animals. Destruction of germinal epithelium of seminiferous tubules by gossypol has been reported in both rams and bulls fed diets containing gossypol [3,4]. According to [5,6], extensive damage to germinal epithelium may be responsible for depression in sperm production, motility, increased sperm abnormalities and depression in spermatogenesis.

A number of studies have been conducted to either reduce or inactivate free gossypol and a number of methods have been proposed: solvent extraction by liquid cyclone and/or acetone [7]; addition of iron salts to cottonseed meal-based diets to bind gossypol [8] and use of calcium hydroxide [9]. Unfortunately, some of these methods had negative effect on protein quality, failed to prevent absorption of free gossypol [10] and increased overall feed cost. Also, breeding of varieties of cotton plant with lower levels of gossypol content and physical extraction of gossypol have also been investigated [11].

The reduction of free gossypol using solvents comes with the difficulty of totally removing residual solvents that may be potentially harmful to the animals that consume them, while calcium hydroxide often reduces the biological activity of vitamins and lowers detoxification efficiency [12]. According to [12], microbial fermentation by Aspergillus niger could tremendously reduce gossypol levels in CSC without any residual effect.

It has been reported that antioxidants play important role in inhibiting the adverse effects of gossypol on cell membranes [13,14]. In view of this, feeding diets containing CSC to monogastrics requires adequate supplementation with antioxidants. Ginger (Zingiber officinale) is a natural source of antioxidants that could possibly neutralize the negative effect of gossypol in CSC. This study therefore, investigated the possibility of using ginger and microbial fermentation by A. niger to counteract the adverse effect of CSC on the reproductive characteristics of rabbit bucks.

II. MATERIAL AND METHODS

2.1 Experimental site

The experiment was conducted at the rabbit research and production unit of the Teaching and Research Farm, Ladoke Akintola University of Technology, (LAUTECH), Ogbomoso (Latitude 8o 151N and Longitude 4o 151E), Oyo State, Nigeria.

2.2 Animals and management

Thirty (30) weaned crossbreed rabbit bucks, New Zealand White X Chinchilla, 6-7 weeks old, were used for the study. Animals (n=6 per treatment) averagely weighing 768.54g were randomly assigned to each of five treatment diets. The bucks were housed individually in wooden metabolic cages. The rabbits were acclimatized for two weeks and afterwards, fed the treatment diets for 12 weeks. Clean drinking water was supplied ad libitum. Feeding was twice per day at 8.00am and 4.00pm, with an allowance of 120g/animal/per day.

2.3 Isolation of organisms and preparation of inoculum

Isolation of organism and preparation of inoculum followed the method of [15]. Aspergilus niger was isolated from CSC using potato dextrose agar (PDA) supplemented with 20% sucrose. Stock culture was maintained on media after serial dilution. The strain was bred and obtained from the research laboratory of Pure and Applied Biology, LAUTECH, Ogbomoso. Inoculum medium was sterilized and incubated at 30±10C for 48hrs. Inoculum was developed by transferring a loopful of mycelium into the inoculum

medium (1% sucrose, 0.2% yeast extract, pH 5.50). The inoculated bottles were incubated at 30±10C on a rotary shaker at 100rev/min for 24hrs. The harvested suspension was stored in the refrigerator until used.

2.4 Preparation of solid substrate

The initial moisture content of dried CSC was determined by drying to constant weight at 1100C in a hot-air oven. Sterilization was carried out in a "fermenter" with 24kg carrying capacity of the substrate. Moisture content was raised to 60%. The CSC was sterilized with the fermenter by autoclaving at 1210C for 3hr and allowed to cool, inoculated with the inoculum and left to ferment for 7 days at 30±10C. Prior to autoclaving, each tray containing CSC was covered with muslin cloth and aluminum foil. The substrate was supplemented with sucrose and yeast extract to enhance microbial growth.

2.5 Preparation of experimental diets

After fermentation, the substrate was dried to constant weight and tagged fermented CSC. A portion of unfermented CSC was kept as raw CSC. The raw CSC and fermented CSC were used in preparation of experimental diets. Also, raw ginger was processed into powder form and used to supplement the diets at 30mg/kg according to recommended dose [16]. Five experimental diets were formulated such that the Control had soybean meal (SBM) as the main protein source. Four other diets were formulated by replacing SBM with either raw or fermented CSC weight for weight, with or without ginger supplementation.

2.6 Experimental Design

The experiment was laid out in a 2X2 factorial arrangement within the Completely Randomized Design (CRD).

2.7 Proximate Analysis

Samples of experimental diets, raw and fermented CSC were analyzed for proximate composition according to [17]

2.8 Reproductive Characteristics

At the end of the feeding trial, animals were slaughtered and reproductive organs (testis and epididymis) were carefully dissected out for evaluation. Data were obtained for the following parameters: Testicular sperm count, Daily Sperm Production (DSP), Sperm morphology, Gonadal Sperm Production (GSP) and Reserve (GSR), and Extra - Gonadal Sperm Reserve (EGSR).

2.9 Determination of Sperm Morphology

Sperm morphology was determined according to the method of [18]. A smear of the semen samples was made by cutting the left testis along the equatorial region and rubbing the cut surface on a clear glass slide. Two drops of eosin-nigrosin dye that had been thoroughly mixed were added. The slides were viewed under a light microscope to identify normal and abnormal cells from several fields. The normal cells were then expressed as the percentage of number of cells counted on each field of the slide. Dead cells were also identified and recorded.

2.10 Gonadal Sperm Production (GSP) and Gonadal Sperm Reserve (GSR)

Gonadal sperm reserve and gonadal sperm production were determined haemocytometrically by homogenate technique using a modification of the method of [19, 20] as described by [14,21]. The tunica

albuginea was removed from the testis and the testicular parenchyma was weighed. A portion of the parenchyma tissue was homogenized by maceration for about 5 minutes in a beaker containing 10 ml physiological saline solution. The homogenate was filtered through a double layer of cheese cloth and the filtrate diluted to ratio 1:20 with de-ionized water. Some drops of the homogenate were introduced into an improved Neubauer haemocytometer counting chamber. All the elongated spermatids and mature sperm cells in the four diagonal and the centre squares of the haemocytometer were counted in each diluted homogenate. The concentration of the sperm cells per gram of testis parenchyma was calculated as follows:

$$\text{Concentration per gram} = \frac{\text{No of sperm cells x volume used}}{\text{Weight of the sample}}$$

Gonadal sperm reserve was calculated as: Concentration / gram testis X total weight of right testis.

2.12 Daily Sperm Production (DSP)

Daily sperm production (DSP) was determined from gonadal sperm reserve (GSR). The daily sperm production was obtained using the formula proposed by [21].

$$\textbf{DSP} = \frac{\text{Testis sperm count (GSR)}}{\text{Time divisor}}$$

The value of time divisor for rabbits = 3.43 [22]. Daily sperm production per gram parenchyma (testis) per animal was estimated by the formula:

$$\text{DSP/g} = \frac{\text{Gonadal Sperm Reserve}}{(\text{Testis wt} - \text{Tunica albuginea wt}) \times 3.43}$$

2.13 Extra - Gonadal Sperm Reserve (EGSR)

Samples of cauda epididymis were homogenized separately in normal saline solution at 100mg/ml and then filtered through a double layer cheese cloth. The resulting homogenate was diluted with de-ionized water at 25 x dilution factor. The sperm count was determined using the improved Neubauer haemocytometer [20, 23].

2.14 Statistical analysis

Data obtained were subjected to one-way analysis of variance (ANOVA) and General Linear Model appropriate for a 2 X 2 factorial experiment, using [24]. Means were separated by Duncan`s multiple range option of the same statistical package.

III. RESULTS AND DISCUSSION

3.1 Results

Sperm characteristics of rabbit bucks fed raw CSC or fermented CSC with or without ginger supplementation is presented in Table 2. Sperm concentration, motile sperm, dead sperm, GSR and EGSR were significantly ($P<0.05$) affected by the treatment. Abnormal sperm and daily sperm production/gram testis were not significantly ($P>0.05$) affected by dietary treatments. The sperm concentration was significantly ($P<0.05$) higher for the ginger supplemented groups than the control and the non-supplemented groups (41.00 and 43.33x106). Motile sperm were significantly ($P<.0.05$) lowered for animals that fed on raw CSC with ginger compared with bucks fed fermented CSC plus

ginger and the control. Dead sperm was significantly (P<0.05) reduced by ginger supplementation for either raw or fermented CSC. Highest dead sperm count was observed for bucks that were fed raw CSC without ginger supplementation.

Gonadal sperm reserve (GSR) in bucks fed fermented CSC with ginger supplementation was significantly (P<0.05) higher than other treatments including the control. The GSR tended to be lower in nonsupplemented groups compared to the supplemented. Extra-gonadal sperm reserve (EGSR) was significantly (P<0.05) lower in bucks fed raw CSC with or without ginger supplementation than their counterparts fed with fermented CSC with or without ginger. The bucks that were exposed to fermented CSC with ginger supplementation had higher (p<0.05) EGSR than the control group. Daily sperm production (DSP) was significantly (P<0.05) higher in supplemented groups than their non-supplemented counterparts.

Table 3 shows the main effect of cottonseed cake on sperm characteristics of rabbit bucks. Significant (p<0.05) variations were recorded between raw and fermented CSC groups for sperm concentration, dead sperm and EGSR. Sperm concentration for bucks that fed on fermented CSC was 13.5% higher than that of the bucks that fed on raw CSC diet. Inclusion of fermented CSC in diet of rabbit bucks resulted in 52.3% increase in extra-gonadal sperm reserve.

Table 4 shows the effect of ginger supplementation on sperm characteristics of rabbit bucks. Bucks that were fed ginger-supplemented diets had significantly higher sperm concentration than the nonsupplemented groups. Feeding bucks with gingersupplemented diet resulted in significantly (p<0.05) higher GSR, EGSR and DSP than the nonsupplemented. Ginger supplementation had no significant effect on motile sperm and abnormal sperm.

3.2 Discussion

The significant increase observed for sperm concentration in the supplemented groups could be attributed to the action of ginger which probably inhibited the anti-fertility effect of gossypol on sperm cells. Ginger has been reported to possess antioxidant activities due to the presence of gingerol-related compounds and diarylheptanoids [25]. Probably these antioxidant activities of ginger worked against the activities of gossypol on the seminiferous tubules of the bucks. It has been known that one of the mechanisms of action of gossypol involves generation of reactive oxygen species (ROS) which are deleterious by-products of metabolism.

The observation that a combination of fermentation and ginger supplementation tended to increase the sperm concentration more than the raw CSC with ginger supplementation is an indication of a synergistic action against the adverse effects of gossypol. Fermentation has been shown to lower gossypol level in CSC [12], while ginger, with its antioxidant property [25], might have inhibited formation of ROS by the residual gossypol content in the CSC. These combined action possibly paved the way for unhindered spermatogenesis that resulted in higher sperm concentration. This observation is consistent with the report of [26] that ginger confers a protective antioxidant defense capacity on rats.

The significant reduction observed for motile sperm in bucks fed raw CSC with ginger supplementation could be associated with the strong effect of gossypol in the raw CSC on sperm cells which ginger supplementation alone could not neutralize. Similar observation was reported by [5] who reported a decrease in number and motility of spermatozoa for bulls fed CSC without any supplementation. The

significantly lower DSP observed in bucks fed raw CSC without ginger supplementation could be attributed to the effect of gossypol in the CSC diets which might have caused some damage to the germinal epithelium of the seminiferous tubules leading to lower concentration of cytoplasmic defensive enzymes against lipid peroxidation in the late spermiogenesis [5,27]. This could also be related to the significantly higher value recorded for dead sperm in the bucks fed the same diet. However in the bucks fed with fermented CSC supplemented with ginger, the Z. officinale possibly counteracted the reduction in DSP by inhibiting the damage that might have been caused by reactive oxygen species (ROS) or by sparing the effect of enzymes involved in the antioxidant system of the testicle. Moreover, the possible destruction of germinal epithelial layer of the seminiferous tubules by gossypol was probably inhibited by ginger, demonstrating its antioxidant potential by capturing the oxygen free radicals that might have been generated by gossypol, thereby enhancing spermatogenesis. Also, the significant increase observed in EGSR of bucks fed fermented CSC without ginger and fermented CSC with ginger was directly related to the mechanisms that resulted in high DSP which consequently translated to increased GSR.

CONCLUSION

Findings from this study show that total replacement of SBM with raw CSC reduced sperm quality in rabbit bucks. A combination of fermentation (using Aspergillus niger) and ginger supplementation corrected the adverse effect. Hence, a demonstration of the potential to enhance the utilization of CSC by rabbit bucks without adverse effect on sperm characteristics.

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Ingredients (%)	Control	- Ginger		+ Ginger	
	SBM	Raw CSC	Fermented CSC	Raw CSC	Fermented CSC
Maize	43.57	43.24	43.57	43.24	43.57
Soybean meal	20.43	-	-	-	-
Cottonseed cake	-	20.43	20.43	20.43	20.43
Rice husk	21	21	21	21	21
Brewers dry grain	10.00	10.33	10.00	10.33	10.00
Fishmeal (72%)	2.00	2.00	2.00	2.00	2.00
Oyster shell	2.00	2.00	2.00	2.00	2.00
Bone meal	0.25	0.25	0.25	0.25	0.25
Premix*	0.25	0.25	0.25	0.25	0.25
Salt	0.20	0.20	0.20	0.20	0.20
Lysine	0.10	0.10	0.10	0.10	0.10
Methionine	0.20	0.20	0.20	0.20	0.20
Total	**100.00**	**100.00**	**100.00**	**100.00**	**100.00**
Ginger (mg/kg)		-	-	30	30
Calculated nutrients					
Crude protein (%)	16.32	15.72	16.13	15.72	16.13
Crude Fiber (%)	10.22	11.63	11.57	11.63	11.57
ME (Kcal/kg)	2582.00	2469.00	2436.00	2469.00	2436.00
Lysine	0.86	0.61	0.64	0.61	0.64
Methionine	0.27	0.23	0.24	0.23	0.24

Table 1: Gross composition and calculated nutrients of experimental diets

*Premix composition (per kg of diet): vitamin A, 12,500 IU; vitamin D3, 2500 IU; vitamin E, 50.00K3, 2.50mg; vitamin B1, 3.00mg; vitamin
B2, 6.00mg; vitamin B6, 6.00mg; niacin, 40mg; calcium pantothenate, 10mg; biotion,0.08mg; vitamin B12,0.26mg; folic acid, 1.00mg; chlorine chloride, 300mg; manganese, 100mg; iron, 50mg;zinc,45mg; copper, 2.00mg; iodine, 155mg; cobalt, 0.25mg; selenium, 0.10mg; antioxidant, 200mg. Ginger was added to the diets at 30mg/kg SBM: soybean meal, CSC: cottonseed based diets

Parameter		-Ginger		+Ginger			
	SBM	Raw CSC	Fermented CSC	Raw CSC	Fermented CSC	SEM	CSC X Ginger
Sperm concentration ($x10^6$/ml)	42.00^{b}	41.00^{b}	43.33^{b}	60.00^{a}	69.33^{a}	3.34	NS
Motile sperm (%)	78.17^{a}	70.73^{ab}	71.20^{ab}	66.07^{b}	79.97^{a}	1.77	NS
Abnormal sperm (%)	15.83	22.23	18.60	22.16	15.00	1.47	NS
Dead sperm (%)	6.00^{bc}	10.00^{a}	7.57^{b}	5.70^{c}	4.87^{c}	0.52	NS
GSR ($x10^6$)	52.99^{bc}	48.17^{c}	48.73^{c}	55.55^{bc}	66.44^{a}	3.83	NS
EGSR ($x10^6$)	63.25^{b}	71.33^{b}	91.18^{ab}	60.06^{b}	109.98^{a}	7.47	NS
DSP ($x10^6$)	15.45^{b}	14.05^{b}	14.21^{b}	20.26^{a}	19.37^{a}	1.56	NS
DSP/g ($x10^6$)	10.07	9.01	10.45	12.95	13.51	1.12	NS

Table 2: Sperm characteristics of rabbit bucks fed raw or fermented CSC supplemented with or without ginger supplementation

a,b,c. Means within the same row with different superscrips differ significantly (P<0.05); GSR: Gonadal Sperm Reserve; DSP: Daily Sperm Production; EGSR: Extra Gonadal Sperm Reserve; DSP/g: Daily Sperm Production / gram; SEM: standard error of the means; NS: Interaction not significant.

Parameter	Cottonseed cake		
	Raw	Fermented	SEM
Sperm concentration ($x10^6$ /ml)	50.50^b	57.33^a	2.65
Motile sperm (%)	69.90	75.05	2.65
Abnormal sperm (%)	20.32	18.62	2.90
Dead sperm (%)	7.85^a	6.23^b	0.36
GSR ($x10^6$)	51.86	57.58	5.58
EGSR ($x10^6$)	65.69^b	100.58^a	10.06
DSP ($x10^6$)	16.15	18.79	2.52
DSP/g ($x10^6$)	10.78	11.98	1.79

Table 3: Effect of cottonseed cake on sperm characteristics of rabbit bucks

a,b: Means within the same row with different superscrips differ significantly (P<0.05); GSR: Gonadal Sperm Reserve; DSP: Daily Sperm Production; EGSR: Extra Gonadal Sperm Reserve; DSP/g: Daily Sperm Production / gram; SEM: standard error of the means.

Parameter	Ginger		
	Minus (-)	Plus (+)	SEM
Sperm concentration ($x10^6$ /ml)	43.18^b	64.67^a	3.04
Motile sperm (%)	70.97	73.02	2.40
Abnormal sperm (%)	20.42	18.52	4.11
Dead sperm (%)	8.78^a	5.28^b	0.50
GSR ($x10^6$)	6.62^b	10.31^a	1.22
EGSR ($x10^6$)	6.29^b	13.88^a	3.70
DSP ($x10^6$)	15.80^b	19.10^a	0.68
DSP/g ($x10^6$)	2.63	2.90	1.33

Table 4: Effect of ginger supplementation on sperm characteristics of rabbit bucks

a,b: Means within the same row with different superscrips differ significantly (P<0.05); GSR: Gonadal Sperm Reserve; DSP: Daily Sperm Production; EGSR: Extra Gonadal Sperm Reserve; DSP/g: Daily Sperm Production / gram; SEM: standard error of the means.

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