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Volume 17 Number 7, July 2018

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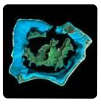
Original Research Articles



Influence of drug-polymer ratio on physical characteristics and release of metformin hydrochloride from metformin-alginate microspheres HTML (abstract.php?id=2183&aTitle=Influence of drug-polymer ratio on physical characteristics and release of metformin hydrochloride from metformin-alginate microspheres) | Fulltext (../admin/12389900798187/2018_17_7_1.pdf)

Dewi Melani Hariyadi (mailto:dewi-m-h@ff.unair.ac.id), *Esti Hendradi*, *Tristiana Erawati*, *Edlin Nur Jannah*, *Wenny Febrina*,
http://dx.doi.org/10.4314/tjpr.v17i7.1 (http://dx.doi.org/10.4314/tjpr.v17i7.1)



Enhanced production of butyric acid by solid-state fermentation of rice polishings by a mutant strain of Clostridium tyrobutyricum HTML (abstract.php?id=2184&aTitle=Enhanced production of butyric acid by solid-state fermentation of rice polishings by a mutant strain of Clostridium tyrobutyricum) | Fulltext (../admin/12389900798187/2018_17_7_2.pdf)

Tasleem Akhtar (mailto:tasleem_ak@yahoo.com), *Abu Saeed Hashmi*, *Muhammad Tayyab*, *Aftab Ahmed Anjum*, *Shagufta Saeed*,
http://dx.doi.org/10.4314/tjpr.v17i7.2 (http://dx.doi.org/10.4314/tjpr.v17i7.2)



Diosgenin inhibits cell proliferation of primary human thyrocytes via downregulation of PI3K/Akt signaling pathway HTML (abstract.php?id=2185&aTitle=Diosgenin inhibits cell proliferation of primary human thyrocytes via downregulation of PI3K/Akt signaling pathway) | Fulltext (../admin/12389900798187/2018_17_7_3.pdf)

Fen Wen, *Yuxian Lu*, *Ke Xu*, *Yanmei Liu*, *Xiaojuan Qian*, *Dezhi Bian* (mailto:dezhiBian2008@163.com),
http://dx.doi.org/10.4314/tjpr.v17i7.3 (http://dx.doi.org/10.4314/tjpr.v17i7.3)



Suppression of long non-coding RNA H19 inhibits proliferation, cell migration and invasion in human cervical cancer cells HTML (abstract.php?id=2186&aTitle=Suppression of long non-coding RNA H19 inhibits proliferation, cell migration and invasion in human cervical cancer cells) | Fulltext (../admin/12389900798187/2018_17_7_4.pdf)

Huawei Xin, *Mingzhe Li*, *Xiaoling Cheng*, *Tao Wang*, *Xiaoliu Liu*, *Yan Zhang* (mailto:IzettaNnna@yahoo.com),
http://dx.doi.org/10.4314/tjpr.v17i7.4 (http://dx.doi.org/10.4314/tjpr.v17i7.4)



Peptide 17, an inhibitor of YAP/TEAD4 pathway, mitigates lung cancer malignancy HTML (abstract.php?id=2187&aTitle=Peptide 17, an inhibitor of YAP/TEAD4 pathway, mitigates lung cancer malignancy) | Fulltext (../admin/12389900798187/2018_17_7_5.pdf)

Jirong Zhang, *Yong Pan*, *Dehua Liao*, *Jingyi Tang*, *Dunwu Yao* (mailto:yaodunwumedchs@163.com),
http://dx.doi.org/10.4314/tjpr.v17i7.5 (http://dx.doi.org/10.4314/tjpr.v17i7.5)



Characteristics and anticancer properties of Sunitinib malate-loaded poly-lactic-co-glycolic acid nanoparticles against human colon cancer HT-29 cells lines HTML (abstract.php?

id=2188&aTitle=Characteristics and anticancer properties of Sunitinib malate-loaded poly-lactic-co-glycolic acid nanoparticles against human colon cancer HT-29 cells lines) | Fulltext (../admin/12389900798187/2018_17_7_6.pdf)

Abdullah S Alshetaili (mailto:a.alshetaili@psau.edu.sa), *Md Khalid Anwer*, *Saad M Alshahrani*, *Ahmed Alalaiwe*, *Bader B Alsulays*, *Mohammad Javed Ansari*, *Faisal Imam*, *Sultan Alshehri*,

http://dx.doi.org/10.4314/tjpr.v17i7.6 (http://dx.doi.org/10.4314/tjpr.v17i7.6)



Apoptosis-inducing effect of 6,7-dimethoxy-4-hydroxy-8-formylflavon from Nicotiana tabacum L leaf in human hepatoma HepG2 cells via activation of mitochondria-mediated apoptotic pathway HTML (abstract.php?id=2189&aTitle=Apoptosis-inducing effect of 6,7-dimethoxy-4-hydroxy-8-formylflavon from Nicotiana tabacum L leaf in human hepatoma HepG2 cells via activation of mitochondria-mediated apoptotic pathway) | Fulltext (../admin/12389900798187/2018_17_7_7.pdf)

Qiu-Jie Zhang, *Wen-Sheng Qiu*, *Hong-Xia Cui*, *Zhuang Yu*, *Ru-Yong Yao*, *Shi-Hai Liu*, *Zi-Min Liu*, *Jun Liang* (mailto:junlianginqd@163.com),

http://dx.doi.org/10.4314/tjpr.v17i7.7 (http://dx.doi.org/10.4314/tjpr.v17i7.7)



Indole-3-acetate induces apoptosis and stimulates phosphorylation of p65NF-KB in 143B and HOS osteosarcoma cells HTML (abstract.php?id=2190&aTitle=Indole-3-acetate induces apoptosis and stimulates phosphorylation of p65NF-KB in 143B and HOS osteosarcoma cells) | Fulltext (../admin/12389900798187/2018_17_7_8.pdf)

Yanhui Zhang, *Yang Li*, *Chao Wang*, *Kewen Zheng*, *Chen Feng*, *Wenbo Wang* (mailto:WadsworthCbeish@yahoo.com),

http://dx.doi.org/10.4314/tjpr.v17i7.8 (http://dx.doi.org/10.4314/tjpr.v17i7.8)



Potential application of Conyza canadensis (L) Cronquist in the management of diabetes: In vitro and in vivo evaluation HTML (abstract.php?id=2191&aTitle=Potential application of Conyza canadensis (L) Cronquist in the management of diabetes: In vitro and in vivo evaluation) | Fulltext (../admin/12389900798187/2018_17_7_9.pdf)

Huma Aslam, *Arif-ullah Khan* (mailto:arif.ullah@riphah.edu.pk), *Humaira Naureen*, *Fawad Ali*, *Farhat Ullah*, *Abdul Sadiq*,

http://dx.doi.org/10.4314/tjpr.v17i7.9 (http://dx.doi.org/10.4314/tjpr.v17i7.9)



Studies on intestinal passage of flumequine and oxytetracycline-loaded MIL-100 (Fe) in the presence of divalent ions HTML (abstract.php?id=2192&aTitle=Studies on intestinal passage of flumequine and oxytetracycline-loaded MIL-100 (Fe) in the presence of divalent ions) | Fulltext (../admin/12389900798187/2018_17_7_10.pdf)

Fatma Ben Ayed (mailto:go.mad289@gmail.com), *Godefroy Mamadou*, *Hanae Naceiri Mrabti*, *Nicolas Limas-Nzouzi*, *Bruno Eto*, *Saad Saguem*,

http://dx.doi.org/10.4314/tjpr.v17i7.10
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Phytochemical screening, antioxidant, antiulcer and toxicity studies on Desmodium adscendens (Sw) DC Fabaceae leaf and stem HTML (abstract.php?id=2193&aTitle=Phytochemical screening, antioxidant, antiulcer and toxicity studies on Desmodium adscendens (Sw) DC Fabaceae leaf and stem) | Fulltext (../admin/12389900798187/2018_17_7_11.pdf)

Gloria A Ayoola, *Samuel O Eze*, *Oluwatosin O Johnson* (mailto:tosyn.villa@gmail.com), *David K Adeyemi*,

http://dx.doi.org/10.4314/tjpr.v17i7.11
(http://dx.doi.org/10.4314/tjpr.v17i7.11)



Possible role of 18-kDa translocator protein (TSPO) in etifoxine-induced reduction of direct twitch responses in isolated rat nerve-skeletal muscle preparations HTML (abstract.php?id=2194&aTitle=Possible role of 18-kDa translocator protein (TSPO) in etifoxine-induced reduction of direct twitch responses in isolated rat nerve-skeletal muscle preparations) | Fulltext (../admin/12389900798187/2018_17_7_12.pdf)

Plamen I Zagorchev, *Vesela Yu Kokova* (mailto:vesela_uk@abv.bg), *Elisaveta G Apostolova*, *Lyudmil P Peychev*,
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(http://dx.doi.org/10.4314/tjpr.v17i7.12)



Purification, compositional analysis and antioxidant properties of polysaccharides from black ginseng HTML (abstract.php?id=2195&aTitle=Purification, compositional analysis and antioxidant properties of polysaccharides from black ginseng) | Fulltext (../admin/12389900798187/2018_17_7_13.pdf)

Li-Hong Gong, *Tao Lei*, *Zhao-Li Zhang*, *Qi-Chao Liang*, *Feng-Guo Zhai*, *Yi-Yan Wu*, *Xiu-Ping Zhang*, *Jia-Qi Liu*, *Jia-Wei Liu* (mailto:jwliu1985@163.com),
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Effect of maternal diabetes and quercetin exposure on the oxidative stress and kidney damage in rat fetus HTML (abstract.php?id=2196&aTitle=Effect of maternal diabetes and quercetin exposure on the oxidative stress and kidney damage in rat fetus) | Fulltext (../admin/12389900798187/2018_17_7_14.pdf)

Saeedeh Heidarinazhad, *Mahmood Khaksary Mahabady* (mailto:mkhaksary@scu.ac.ir), *Reza Ranjbar*, *Hossein Najafzadeh Varzi*, *Babak Mohammadian*, *Mohammad Reza Tabandeh*,
http://dx.doi.org/10.4314/tjpr.v17i7.14
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Testicular morphology and seminal fluid parameters of adult Wistar rats following honey administration HTML (abstract.php?id=2197&aTitle=Testicular morphology and seminal fluid parameters of adult Wistar rats following honey administration) | Fulltext (../admin/12389900798187/2018_17_7_15.pdf)

Eniola R Kadir (mailto:ennyshittabey@yahoo.com), *Lekan S Ojulari*, *Abdulmumin Ibrahim*, *Oluwole J Ekundayo*, *Rukayat Jaji-Sulaimon*, *Hidaayah Jimoh-Abdulghaffaar*,
Honey, Fertility, Spermatozo (Honey, Fertility, Spermatozo)



Antitumor effects of candidone extracted from Derris indica (Lamk) Bennet in cholangiocarcinoma cells HTML (abstract.php?id=2198&aTitle=Antitumor effects of candidone extracted from Derris indica (Lamk) Bennet in cholangiocarcinoma cells) | Fulltext (../admin/12389900798187/2018_17_7_16.pdf)

Benjawan Kurasug, *Veerapol Kukongviriyapan*, *Auemduan Prawan*, *Chavi Yenjai*, *Sarinya Kongpetch* (mailto:sarinyako@kku.ac.th),
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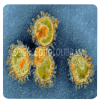

Indonesian Pasuruan propolis extract does not exert anti-proliferation and pro-apoptotic effect on human colon carcinoma cell line HT-29 HTML (abstract.php?id=2199&aTitle=Indonesian Pasuruan propolis extract does not exert anti-proliferation and pro-apoptotic effect on human colon carcinoma cell line HT-29) | Fulltext (../admin/12389900798187/2018_17_7_17.pdf)

Ihda Dian Kusuma (mailto:ihdadiankusuma@ymail.com), *Aris Rosidah*, *Eviana Norahmawati*, *Nurdiana* , *Agustina Tri Endharti*,

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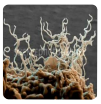

Role of OCT4 in cisplatin treatment of testicular embryonal carcinoma HTML (abstract.php?id=2200&aTitle=Role of OCT4 in cisplatin treatment of testicular embryonal carcinoma) | Fulltext (../admin/12389900798187/2018_17_7_18.pdf)

Qin Le, *Jie Lin*, *Xiaoxiao Xie*, *Xiangbo Yu*, *Yang Cai*, *Yangping Shentu*, *Aihua Zhang* (mailto:zhangaihuaxwsdoc@163.com), *Aiwu Li* (mailto:liaiwuxwymed@163.com),
http://dx.doi.org/10.4314/tjpr.v17i7.18
(http://dx.doi.org/10.4314/tjpr.v17i7.18)



Effect of dexmedetomidine hydrochloride combination with conventional anesthesia on serum cortisol, inflammatory factors and cellular immunity during surgery on children with congenital anus atresia HTML (abstract.php?id=2201&aTitle=Effect of dexmedetomidine hydrochloride combination with conventional anesthesia on serum cortisol, inflammatory factors and cellular immunity during surgery on children with congenital anus atresia) | Fulltext (../admin/12389900798187/2018_17_7_19.pdf)

Aihua Liu, *Lichuan Tian*, *Fang Yin* (mailto:jn1336@163.com),
http://dx.doi.org/10.4314/tjpr.v17i7.19
(http://dx.doi.org/10.4314/tjpr.v17i7.19)



Effect of hydroalcohol extract of lemon (Citrus limon) peel on a rat model of type 2 diabetes HTML (abstract.php?id=2202&aTitle=Effect of hydroalcohol extract of lemon (Citrus limon) peel on a rat model of type 2 diabetes) | Fulltext (../admin/12389900798187/2018_17_7_20.pdf)

Juan Lv, *Lanxiu Cao*, *Min Li*, *Rui Zhang*, *Fu Bai*, *Pengfei Wei*,
http://dx.doi.org/10.4314/tjpr.v17i7.20
(http://dx.doi.org/10.4314/tjpr.v17i7.20)



Anti-diabetic activity of aqueous extract of Fructus Ligustri Lucidi in a rat model of type 2 diabetes HTML (abstract.php?id=2203&aTitle=Anti-diabetic activity of aqueous extract of Fructus Ligustri Lucidi in a rat model of type 2 diabetes) | Fulltext (../admin/12389900798187/2018_17_7_21.pdf)

Juan Lv, *Lanxiu Cao* (mailto:lanxiucaodoczhej@126.com), *Rui Zhang*, *Pengfei Wei*,
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Investigation of nedaplatin and CpG oligodeoxynucleotide combination therapy in a mouse model of lung cancer HTML (abstract.php?id=2204&aTitle=Investigation of nedaplatin and CpG oligodeoxynucleotide combination therapy in a mouse model of lung cancer) | Fulltext (../admin/12389900798187/2018_17_7_22.pdf)

Jianfeng Quan, *Yanli Zhao* (mailto:yanli_z@yeah.net),
http://dx.doi.org/10.4314/tjpr.v17i7.22
(http://dx.doi.org/10.4314/tjpr.v17i7.22)



Inhibitory effect of α-cyclodextrin on α-amylase activity HTML (abstract.php?id=2205&aTitle=Inhibitory effect of α-cyclodextrin on α-amylase activity) | Fulltext (../admin/12389900798187/2018_17_7_23.pdf)

Min Wang, *Qingsong Gou*, *Lingyi Liu* (mailto:liulingyi607@zju.edu.cn), *Zhan Wang*, *Zhenzhou Zhu*, *Fang Li*, *Wei Zhang*, *Wangyang Shen*,
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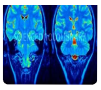

Urinary paraquat concentration and white blood cell count as prognostic factors in paraquat poisoning HTML (abstract.php?id=2206&aTitle=Urinary paraquat concentration and white blood cell count as prognostic factors in paraquat poisoning) | Fulltext (../admin/12389900798187/2018_17_7_24.pdf)

Qinliang Xu, *Xinli Wang*, *Qiang Wu*, *Xiangdong Jian* (mailto:ih0713@163.com), *Baotian Kan*, *Beijun Gao*, *Ke Wang*,
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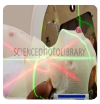

Synthesis of 3-[4-(2-furoyl)-1-piperazinyl]-N-(substituted)propanamides as promising antibacterial agents with mild cytotoxicity HTML (abstract.php?id=2207&aTitle=Synthesis of 3-[4-(2-furoyl)-1-piperazinyl]-N-(substituted)propanamides as promising antibacterial agents with mild cytotoxicity) | Fulltext (../admin/12389900798187/2018_17_7_25.pdf)

Ghulam Hussain, *Muhammad A Abbasi* (mailto:abbasi@gcu.edu.pk), *Aziz-ur-Rehman* , *Sabahat Z Siddiqui*, *Irshad Ahmad*, *Rabia Malik*, *Muhammad Shahid*, *Zahid Mushtaq*, *Syed AA Shah*,
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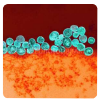

Etiologic analysis of Chinese patients with agranulocytosis and hematopathies infected with resistant bacteria: Anti-bacterial effect of tigecycline HTML (abstract.php?id=2208&aTitle=Etiologic analysis of Chinese patients with agranulocytosis and hematopathies infected with resistant bacteria: Anti-bacterial effect of tigecycline) | Fulltext (../admin/12389900798187/2018_17_7_26.pdf)

Feifei Che (mailto:feifeiche@cntv.cn), *Chunqian Wan*, *Xiaodong Wang*, *Jiao Chen*, *Juan Huang*,
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Perceived causes of prescribing errors by physicians: A qualitative study HTML (abstract.php?id=2209&aTitle=Perceived causes of prescribing errors by physicians: A qualitative study) | Fulltext (../admin/12389900798187/2018_17_7_27.pdf)

Basmah Al-Fageh, *Hisham Aljadhey*, *Mansour A Mahmoud* (mailto:Mammm.99@gmail.com), *Nouf Al-Fadel*, *Mohamed Azmi Hassali*, *Bryony Dean Franklin*,
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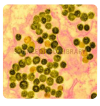

Incidence of adverse drug reactions in a paediatric ward of a Malaysian hospital: A prospective observational study HTML (abstract.php?id=2210&aTitle=Incidence of adverse drug reactions in a paediatric ward of a Malaysian hospital: A prospective observational study) | Fulltext (../admin/12389900798187/2018_17_7_28.pdf)

Muslimah Ithnin, *Mohd Dzulkhairi Mohd Rani*, *Zuraidah Abd Latif*, *Paveethra a/p Kani*, *Asmalita Syaiful*, *Tengku Amatullah Madeehah Tengku Mohd*, *Khairun Nain Nor Aripin* (mailto:khairun@usim.edu.my),
http://dx.doi.org/10.4314/tjpr.v17i7.28
(http://dx.doi.org/10.4314/tjpr.v17i7.28)



Changes in the expressions of E-selectin, adiponectin and serum ferritin in patients with diabetic retinopathy, and their correlations HTML (abstract.php?id=2211&aTitle=Changes in the ex<x>pressions of E-selectin, adiponectin and serum ferritin in patients with diabetic retinopathy, and their correlations) | Fulltext (../admin/12389900798187/2018_17_7_29.pdf)

Zhou Li (mailto:vb1336@163.com), *Zhou Zheng*, *Wei Xiaodan*, *Tian Bin*, *Ji Yuanhong*,
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Dispensing patterns of antimigraine agents with a focus on seasonal variations in prescribing HTML (abstract.php?id=2212&aTitle=Dispensing patterns of antimigraine agents with a focus on seasonal variations in prescribing) | Fulltext (../admin/12389900798187/2018_17_7_30.pdf)

Ilse Truter (mailto:ilse.truter@mandela.ac.za), *Bernadette Louwrens*, *Theunis J van W Kotze*,
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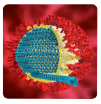
Review Articles



Application of statins in management of glioma: Recent advances HTML (abstract.php?id=2213&aTitle=Application of statins in management of glioma: Recent advances) | Fulltext (../admin/12389900798187/2018_17_7_31.pdf)

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Archives

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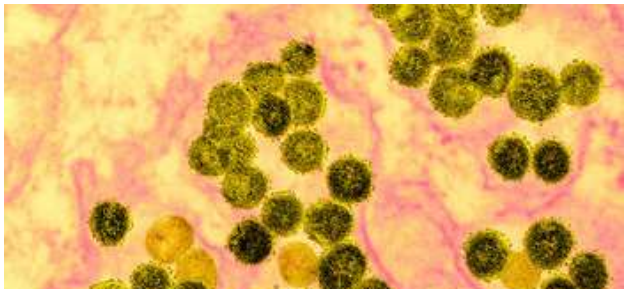

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Original Research Article

Testicular morphology and seminal fluid parameters of adult Wistar rats following honey administration

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Abstract

Purpose: *Honey has a long history of use in the traditional medical systems This objective of this study was to find out the effects of honey on quality and quantity of sperm and testicular microstructure when compared to fertility boosting drug and controls.*
Methods: *A total number of thirty (30) matured male Wistar rats that were sexually active weighing 200 - 280g were used for this study. The animals were grouped into five as A - E. Group A was the control; Group B (standard group) was the standard group that received 0.3ml of follicle stimulating hormone (FSH) drug for 6 days; groups C, D and E received 1ml, 2ml, and 2.5ml of honey daily for 21days respectively. After 21 days of administration, the testes were removed for analysis of the sperm parameters and the histology.*
Results: *Honey significantly improved the sperm quality and spermatogenesis rate (denser seminiferous tubule lumen) of exposed animals compared to control animals, but most improvement was seen in the standard group that received 0.3ml FSH. Also no sign of degeneration or cellular loss was observable in the testicular histo-architecture of experimental animals.*
Conclusion: *This research showed that honey possesses some fertility boosting properties in exposed animals compared to controls and honey is not associated with increased sperm abnormalities.*

Keywords: *Honey, Fertility booster, Spermatozoa, Ologospermia*

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INTRODUCTION

Fertility in males is largely dependent on adequate production of healthy spermatozoa by the testes, these days many men have resolved to the use of alternative substances (like fruits, herbs, honey etc.) for fertility boost [1]. It has been recorded that approximately 70 % of men worldwide use alternative medicinal substances that are cheap and readily available to increase their sperm content [1]. The World health organization [2] endorsed honey and other sperm boosting supplement as being safe for usage [3]. Studies have shown that honey increased spermatogenesis in rats i.e. sperm boosting properties [4]. A report from Malawi suggested that long term administration of honey had no negative effect on the histopathology of Wistar rats' testis [5], and Igbokwe *et al* [6] found increase in sperm parameters in honey treated

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animals, while a contrary result was obtained by Dare *et al* [3] who reported reduced motility and increased percentage of dead and abnormal spermatozoa in honey treated animals.

Bees produce this sweet food (honey) by using nectars from flowers. Honey has a long history of use in the traditional medical systems [7]. It has a density of about 1.36 kg/litre (i.e. 36 % denser than water). The major active constituent which has been found to have strong fertility effect is the Chrysin [8] which is a bioflavonoid compound. This compound is in high quantity in honey and propolis. Chrysin is also known for its testosterone boosting activity Dhawan *et al* [9]. It also inhibits the conversion of testosterone to estrogen [10]. It appears to be poorly absorbed and also sufficiently metabolized, hence low levels are found in testes and blood. These levels are insufficient to exert the beneficial effects of testosterone boosting Kohut *et al* [11].

Chrysin, like most bioflavonoids, is poorly bioavailable in its isolated form. This makes its actual use to be decreased [12]. Its bioavailability is thought to be due to its extensive metabolism and intestinal transportation.

Chrysin, similar to most bioflavonoids, is poorly bioavailable in its isolated form. This makes its actual use to be decreased [12]. Its poor bioavailability is thought to be due to its extensive metabolism and intestinal transportation. Organs containing the P450 system such as the liver, easily conjugates chrysin into chrysin glucuronidase and chrysin sulphate, which are its metabolites though may not be bioactive. Glucuronidation rates are increased in the intestinal cells via induction of UGT1A1 expression by chrysin, which further supports its low bioactive levels [12]. Therefore, this work aims to determine the effect of honey on seminal fluid parameters and morphometry of the seminiferous tubules.

METHODS

Thirty (30) male Wistar rats weighing between 200g-280g were used for the study. They were kept in the animal house of the College of Health Sciences, University of Ilorin under normal atmospheric condition of 25 ^{o}C. The animals were divided into five (5) groups randomly. Each group consist of six (6) rats. Group A was the control; group B was administered 0.3ml of follicle stimulating hormone drug for 6 days [13] (standard group), group C, D and E received 1ml, 2ml, and 2.5ml of honey daily for 21days respectively (increasing quantity of administered honey) [10]

Semen analysis

Sample collection

The Wistar rats were sacrificed after the experiment. They were administered inhalation anaesthetic (ether), then sacrificed by cervical dislocation.

Open castration method was used for orchidectomy via a midline scrotal incision. Then the testicles were gently milked out through the incision site. The testicles were exposed by incising the tunica vaginalis, followed by exposure, ligation and incision of the spermatic cord. The cauda epididymis was then identified, from which the semen samples were collected. Saba *et al* [14] and Oyeyemi and Ubiogoro [15] described a similar method of semen collection. The semen samples were then analysed immediately.

Sperm motility and count assay

A drop of semen was placed on a glass slide to assess sperm motility. 2.9% sodium citrate was added. This was covered with a glass slip, then the microscopic fields were viewed under the light microscope to determine the motile and non-motile spermatozoa. This method was as described by Zemjanis [16].

Sperm count was with the use of a hemocytometer. The cauda epididymis was minced in normal saline, then filtered and an aliquot of 10ul was placed on the hemocytometer. The improved Neubauer chamber (Deep 1/10m, LABART, Munich, Germany) was used for the count, under a light microscope [17].

Sperm morphology and viability assay

Four hundred (400) spermatozoa count were used to determine this. The semen was placed on the slide, then Wells and Awa stains (which consist of 0.2g eosin and 0.6g fast green dissolved in distilled water and ethanol in ratio 2:1) was added, as documented by Wells and Awa [18]; Isaac and Ebenezer [19]. This was then viewed under the microscope

In determining the viability, a drop of semen was placed on the slide, then addition of a drop of eosin-nigrosin stain (which is 1% eosin with 5% nigrosin in 3% sodium citrate solution). This was mixed well then air dried. It was then observed under the microscope for the live and dead spermatozoa cells, which were counted as

described by Wells and Awa [18] and Isaac and Ebenezer [19].

Statistical analysis

Data collected were analyzed using one-way analysis of variance (ANOVA) followed by Tukey's multiple comparison test with the aid of SPSS (V20; USA). The data were presented as means ± SEM (standard error of mean). $P \leq 0.05$ was considered statistically significant.

RESULTS

Histology of the testes

The proliferation of spermatogonic series (SG) into matured spermatozoa in the seminiferous tubular lumen (L) was observed in the control rats (Figure 1). There was an improvement in the proliferation rate of the spermatogenic series (SG) in the FSH treated rats (Figure 2), shown by the presence of abundant spermatozoa in the seminiferous tubular lumen (L), i.e., a dense lumen. The basement membrane was also intact and there were Leydig cells present in the interstitium (I).

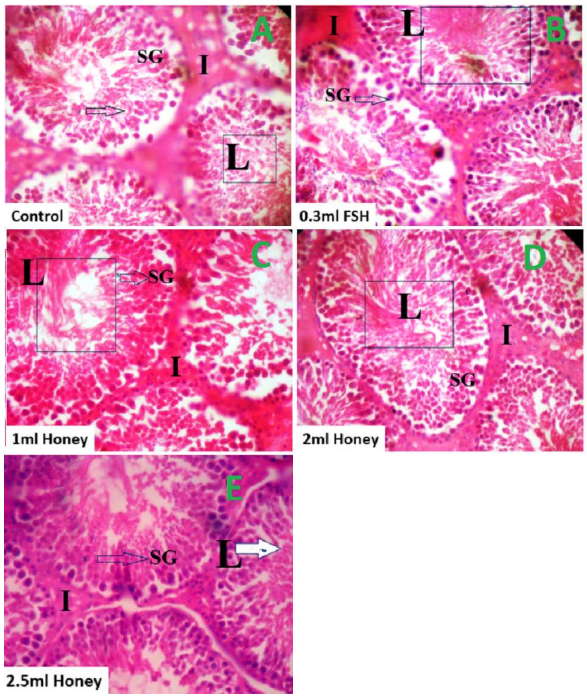

Figure 1: Phoomicrograph showing sections of the seminiferous tubules of control rats, L-lumen (A), rats treated with 0.3ml FSH, L-densely packed lumen (B), rats treated with 1 ml honey, L-densely packed lumen (C), rats treated with 2 ml honey, L-densely packed lumen (D) and rats treated with 2.5 ml honey, L-lumen. Stain H&E; X100. SG – spermatogonia cells, I-intestinal space

Additional increases in the doses of honey in the honey treated groups (III, IV and V) showed progressive proliferation of spermatogonic series (SG) into matured spermatozoa in the seminiferous tubular lumen (L) with intact basement membrane and interstitial space (I). This is shown in Figure 1 (C-E).

Semen analysis

Sperm Count: The sperm count was significantly increased (p<0.001) in the FSH treated group (254.6±4.07 × 10^4/mL) when compared to control (142.2±21.25 × 10^4/mL) (Table 1). A dose dependent significant increase (p<0.01) was also recorded in the honey treated groups (group III-190.90±9.41 × 10^4/mL, group IV- 204.30±4.05× 10^4/mL and group V- 205.33±1.48 × 10^4/mL).

Sperm Motility (%): The sperm motility was significantly increased in all experimental groups when compared to control (66.30±0.99 %), with the highest increase recorded in the FSH treated group (84.76±0.74 % p<0.001). Sperm motility was (70.94±0.92 % p<0.05) (70.53±1.66 % p<0.05) and (74.80±0.53 % p<0.05) in groups III, IV and V, respectively (Table 1).

Sperm Morphology (%): There was significant increase in group II (79.02 ±0.28 % p<0.01) and group V (78.57±2.67 % p<0.05) when compared to control (72.32±2.01 %). Groups III and IV showed insignificant increases (p>0.05) when compared to control (Table 1).

Life/Death Ratio (%): The life/death ratio was increased in all experimental groups (II-87.24±0.86 %, III- 79.18±1.28 %, IV- 75.38±2.89 and V- 80.59±0.89) when compared to control (81.04±1.29 %), but was only significant (p <0.05) in the FSH treated group (Table 1).

DISCUSSION

The histological evaluation of the actions of honey on the microstructure of the testes revealed less dense packing of spermatogenic cells in control compared to the FSH and honey treated groups. Also, the lumen of control was less densely filled with seminiferous tubular lumen with spermatozoa compared to FSH and honey treated groups. The increase in the abundance of spermatozoa in the lumen is dose dependent among the groups that received varying doses of honey as most spermatozoa i.e. denser lumen, was seen in animals that received 2ml and 2.5ml Honey, which is similar to findings by Nworah *et al* [5].

Table 1: Semen profile

Group	Sperm count (Total No. $\times 10^4$)	Sperm motility (%)	Sperm morphology (%)	Life/death ratio (%)
Control	142.20±21.25	66.30±0.99	72.32±2.01	81.04±1.29
0.3ml FSH	254.60±4.07***	84.76±0.74***	79.02±0.28**	87.24±0.86**
1ml honey	190.90±9.41**	70.94±0.92*	74.74±1.55	79.18±1.28
2ml honey	204.33±4.05***	70.53±1.66*	75.67±2.48	75.37±2.89
2.5ml honey	205.33±1.48***	74.80±0.53**	78.57±2.67*	80.57±0.87

*Sperm count, percentage sperm motility, morphology and life death ratio of animals treated FSH and varying doses of honey. Data represented as mean±SEM. * (P≤0.05), ** (P≤0.01), *** (p≤0.001)*

Previous studies revealed that honey of appropriate dose might enhance the third stage of spermatogenesis in rats [20]. Yousef and Salama [21] observed a similar finding of honey acting as a physiologic modulator for spermatogenic cell proliferation, which influence the cell cycle of the seminiferous epithelium thus, increasing spermatogenesis.

Likewise, follicle stimulating hormone has been tested and confirmed to cause an increase in spermatogenesis [22]

The characteristics measured by semen analysis are some of the factors in semen quality that determine the viability of sperm [23]. There was a significant dose-dependent increase in the percentage sperm motility and sperm count following treatment with honey. This corresponds to findings by Igbokwe *et al* [6], who reported the potency of honey as a fertility boosting agent and Salman *et al* [24], whose findings were also suggestive of the beneficial effects of honey on sperm count.

Honey contains several metals, amongst which is zinc [25]. These findings are suggestive of zinc accumulating in the testis during early spermatogenesis. Therefore, it may be important in DNA synthesis and regulation of spematogonial proliferation [26].

CONCLUSION

Though there is not enough established data concerning the medicinal use of honey in treating human male infertility currently, this study revealed that honey is potentially useful as a fertility boosting agent by improving on the qualitative and quantitative parameters of spermatogenesis in a dose dependent manner.

DECLARATIONS

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Conflict of Interest

No conflict of interest associated with this work.

Contribution of Authors

The authors declare that this work was done by the authors named in this article and all liabilities pertaining to claims relating to the content of this article will be borne by them.

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