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Solvent Based Variations in Yield of Bioactive Extracts from the Sclerotium of Pleurotustuber-regium

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ABSTRACT

Background: The therapeutic effectiveness of herbs and fungi used for medicinal purposes is not only a factor of their bioactive constituents but also a factor of both the extraction solvent and extraction method.

Objective: The objective of this study is to extract and analyze the bioactive components in the sclerotum of Pleurotus tuber-regium using different solvents, as to ascertain the solvent that gives a better yield.

Method: A quantity of 10.0 kg of fresh sclerotia of P. tuber-regium purchased at Zarama Market in Southern Nigeria was washed, peeled and the white inner parts were sliced using a sterilized knife. The sliced samples were dried at room temperature for fourteen days. After grinding, the bioactive components were extracted by weighing 10 g RI VDPSOH LQWR WKUHH ZHOO VWRSSHU ERWWOHV DQG HDFK ZDV H hexane and dichloromethane), while that of soxleth extraction was done in a soxleth apparatus, using ethanol as the VROYHQW 7KH SURFHVV ZDV UHSHDWHG WZLFH DQG WKH FRPELQHG D Two milliliters of the extracts were used for gas chromatographic and mass spectroscopy analysis.

Result: The highest peak on the chromatogram for the methanol extract was observed at 32.644 min., while hexane, dichloromethane and soxhlet extracts had their highest peaks at 31.459 min., 14.254 min. and 18.060 min. respectively. 7KH KLJKHVW ELRDFWLYH FRPSRQHQW LQ PHWKDQRO H[WUDFW ZDV + D PHWKDQH ZLWK D YDOXH RI ZKLOH KH[DQH GLFKORUR WHWUDGHFDPHWK\O %LV HWK\OKH[\O SKWKDODWH DQG 3KWKDOL DQG UHVSHFWLYHO\

Conclusion: Hexane and dichloromethane extracts yielded more bioactive components with better nutriceutical and medicinal properties and may be regarded as better solvents for mushroom and fungi extractions..

Keywords: Mushroom, Bioactive components, Extraction methods, Solvents, extracts

INTRODUCTION

Different plants and fungal materials have been used in traditional medicine to solve varieties of health problems. The effectiveness of various combinations of herbs and other ingredients used by traditional medical practitioners depends on the constituents of the selected plant or fungi materials. The availability of the constituents is a factor of the adopted extraction method. The extraction of a given compound from a substrate depends on both the solubility of the compound and the polarity of the solvent. Modern extraction techniques such as accelerated solvent extraction, VXSHUFULWLFDO ÀXLG H[WUDFWLRQ PLFURZDYH DVVLVWHG H[WUDFW in nutraceuticals extractions from plants. These modern techniques are mainly targeted at decreasing extraction time, ZKLOH LQFUHDVLQJ \LHOG DQG HQKDQFLQJ H[WUDFW¶V TXDOLW\ ZLWK WRWDO SKHQROLF FRQWHQW WRWDO ÀDYRQRLG FRQWHQW DQG DQWLR[LGDQW DFWLYLW\ RSWLPL]LQJ XOWUDVRQLF DVVLVWHG H[WUDFWLRQ XVLQJ UHVSRQVH PLQ HWKDQRO FRQFHQWUDWLRQ DQG VROYHQW WR VDPSOH UDW

2LO H[WUDFWLRQ LV PDLQO\ D GLIIXVLRQ SURFHVV LQ ZKLFK WKH VI
sample material, thereby forming a solution of the oil in the solvent. Though some of these plants and fungal materials
DUH VWLOO YHU\ UHOHYDQW LQ WUDGLWLRQDO PHGLFLQH IRU WUHDV
RI WKHLU LVRODWHV KDYH VKRZQ SRWHQWLDO DQWL LQÀDPPDWRU\ D

Pleurotus tuberregium is a notable saprotroph that produces a food storage sclerotium upon its consumption of
GHFD\LQJ ZRRG > @ ,W LV RQH PXVKURRP WKDW ERWK LWV EDVLGL
WKHLU QXWULWLYH DQG PHGLFLQDO SURSHUWLHV > @ 7KH H[WUDFW
IRU WKH WUHDWPHQW DQG PDQDJHPHQW RI GLVHDVHV UDQJLQJ IURP
KHDGDFKH VWRPDFK SUREOHP FROG DVWKPD IHYHU KLJK EORRG
/H%DXHU > @ UHSRUWHG WKH XVH RI WKLV PXVKURRP DQG LWV H[WU
VRXSV DQG IRU WKH WUHDWPHQW RI KHDUW UHODWHG DLOPHQWV Z
FRXJK DQG REHVLW\ LQ WKH 6RXWKHUQ SDUW *Pleurotus tuberregium* JHQHUDO J>UR@Z V$ VLQG$
DQG $XVWUDOLD > @

2WKHU WKHUDSHXWLF DFWLYLWLHV VXFK DV DQWLWXPRXU LP
hypocholesterolaemic, antihypertensive, antihyperglycaemic, antimicrobial and antiviral activities of *Pleurotus* spp.
KDV DOVR EHHQ UHSRUWHG > @ 7KRXJK WKHVH WKHUDSHXWLF DFWL
fermentation broth, mycelia and fruiting bodies of *Pleurotus* VSS > @ WKH ELRFKHPLFDO PHFKD
GXH SRRU FKDUDFWHUL]DWLRQ DQG LGHQWL¿FDWLRQ RI WKH ELRDFW
bioactive components in the sclerotum of *Pleurotus tuber-regium* using different solvents, to analyze the extracts obtained for
LGHQWL¿FDWLRQ RI WKH ELRDFWLYH FRPSRQHQWV DQG WR DVFHUWD

MATERIALS AND METHODS

Sample collection, preparation and extraction

$ TXDQWLW\ RI NJ RI *Pleurotus tuberregium* VFOHURWLD RI WKH IXQJXV ZDV REWDLQHG IURP =DUDPD 0DUNHW LQ ZDVKHG SHHOHG DQG WKH ZKLWH LQQHU SDUWV ZHUH VOLFHG XVLQ DW URRP WHPSHUDWXUH LQ D GXVW IUHH HQYLURQPHQW IRU D SHULO EOHQGHU LQWR D ¿QH SRZGHU 8VLQJ DQ DQDO\WLFDO ZHLJKLQJ EDO ZHOO VWRSSHU ERWWOHV $ YROXPH RI P/ RI D VSHFL¿F H[WUDFWL DGGHG WR HDFK VWRSSHU ERWWOH 7KH PL[WXUHV ZHUH YLJRURXVO VR[OHWK H[WUDFWLRQ ZDV GRQH LQ D VR[OHWK DSSDUDWXV XVLQJ ZDV FROOHFWHG E\ ¿OWHULQJ LQWR D TXDUW] EHDNHU LQ D IXPH K DOLTXRW FROOHFWHG IURP HDFK H[WUDFWLRQ VROYHQW ZHUH VHSD E\ SDVVLQJ WKURXJK D SDVWXUH SLSHWWH SDFNHG ZLWK VLOLFD JH DLU GULHG WR P/ IRU JDV FKURPDWRJUDSKLF DQDO\VLV

GC-MS analysis of extracts

7KH H[WUDFWV ZHUH DQDO\VHG XVLQJ D FRPELQHG JDV FKURPDWRJU $JLOHQW 7HFK ¿WWHG ZLWK D FDSLOODU\ FROXPQ +3 06 SKH XVLQJ +HOLXP DV D FDUULHU JDV DW LQLWLDO FROXPQ WHPSHUDWX ZDV LQFUHDVHG DW *f& SHU PLQXWHV WR *f& DQG KHOG IRU PLQ ZDV GRQH DW LRQL]DWLRQ HQHUJ\ RI H9 7KH RLO ZDV GLOXWHG DXWRPDWLFDOO\ LQMHFWHG LQWR $JLOHQW 7HFK PRGHO PDVV XVLQJ WKH &KHP 2I¿FH VRIWZDUH DWWDFKHG WRWKH 06 OLEUDU\ FRQ¿UPHG XVLQJ WKH GDWDEDVH R7 7HFKQRORJLHV ,QF 1HZ 7LWXWH RI 6WDQ

RESULTS

7KH &KURPDWRJUDP RI ELRDFWLYH FRPSRQHQWV RI PHWKDQRO KH
sclerotia of *P. tuberregium* DUH VKRZQ LQ)LJXUHV 1-4 UHVSHFWLYHO\ 7KH KLJKHVW SHDN IR
REVHUYHG DW PLQ +H[DQH H[WUDFW KDV LWV KLJKHVW SHDN DW
KDYH WKHLU KLJKHVW SHDNV DW PLQ DQG PLQ UHVSHFWL

7KH UHWHQWLRQ WLPH SHUFHQWDJH FRQFHQWUDWLRQ PROHFXODU
components of methanol, hexane, dichloromethane and soxhlet extracts of the sclerotia of *P. tuberregium* DUH VKRZQ
in Tables 1-4 D5 5 5 7HWUDPHWK\O KH[DK\GUR + D
FRPSRQHQW LQ WKH PHWKDQRO H[WUDFW ZLWK D YDOXH RI IR

Figure 1: & K U R P D W R J U D P R I E L R D F W L Y H F R P S R Q H Q W V R I P. *Pubenegium.* H W K D Q R O H [W U D F

Figure 2: & K U R P D W R J U D P R I E L R D F W L Y H F R P S R Q H Q W V R I P. *Pubenegium.* K H [D Q H H [W U D F

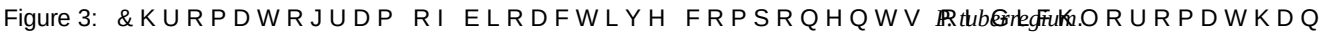


Figure 3: &KURPDWRJUDP RI ELRDFWLYH FRPSRQHQWV IURP *P. tuberregium*.ORURPDWKDQ

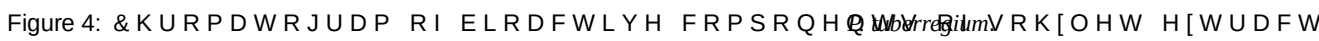


Figure 4: &KURPDWRJUDP RI ELRDFWLYH FRPSRQHQWV IURP *Q. tuberregium*V RK[OHW H[WUDFW

HVWHU EHWD :LWK D YDOXH RI +H[DQH H[WUDFW KDV L
WHWUDGHFDPHWK\O ZLWK D YDOXH RI IROORZHG E\ +H[DFRVL
%LV HWK\OKH[\O SKWKDODWH DQG (LFRVDQH ZHUH WKH KLJKHVW E
ZLWK YDOXHV RI DQG UHVSHFWLYHO\ 7KH KLJKHVW ELRDFW
3KWKDOLF DFLG FKORUREHQ]\O EXW\O HVWHU ZLWK D YDOXH RI
GLPHWKR[\ ZLWK D YDOXH RI

DISCUSSION

The disparity in both compound quantity and concentration observed in the methanol, hexane, dichloromethane and soxhlet extracts of the sclerotia of *P. tuberregium* VKRZV YDU\LQJ FDSDELOLWLHV RI GLIIHU liberate different compounds from a substrate. The ability of methanol to extract only three uncommon polycyclic FRPSRXQGV + &\FORSURSD>D@QDSKWKDOHQ RQH D5 5 5 PHWKDQH DQG 8V HQ RLF DFLG K\GUR[\ PHWK\O HVWHU E 1 indicates that the use of methanol as an extraction solvent may favour the extraction of polycyclic compounds HVSHFLDOO\ LQ D OLJQLQ EDVHG VXEVWUDWH OLNH PXVKURRP $V H[WUDFWV LQ PHWKDQRO H[WUDFWLRQ PD\ EH ORZHU WKDQ LQ D QRQ ODUJHU SURSRUWLRQ RI SRODU FRPSRXQGV +RZHYHU LWV VROXELO polar compounds. The result of this study is an indication that methanol may not be a solvent of choice for extraction ZKHUH ERWK SRODU DQG QRQ SRODU FRPSRXQGV DUH QHHGHG IURP

$OWKRXJK GLIIHUHQW VROYHQWV KDV EHHQ XVHG IRU H[WUDFWLRQ EHFDXVH RI LWV QRQ SRODU SURSHUWLHV ORZ ERLOLQJ SRLQW *&ORXJK DQG 0XOKROODQG > @ DOVR UHSRUWHG WKDW WKH ORZ UHD IRU WKH H[WUDFWLRQ RI UHDFWLYH FRPSRXQGV 7KH KH[DQH H[WUDF GLIIHUHQW ELRDFWLYH FRPSRQHQWV PDGH XS RI OLJQDQ (Table 2). The EUDQFK ODUJH QXPEHU RI FRPSRXQGV REVHUYHG LQ WKH KH[DQH H[WUDFW release these compounds from the substrate.

7KH FRPSRQHQWV REVHUYHG IURP (Table 1) WKH GLFKORURPHWKDQH WR GLVVROYH D ZLGH UDQJH RI RUJDQLF FRPSRX > @ 7KH SUHVHQFH RI ERWK OLQHDU DQG F\FOLF FRPSRXQGV LQ WK potentials to penetrate and extract most constituent compounds from a lignin based sample. Though dichloromethane LV WKH OHDVW WR[LF DPRQJVW WKH VLPSOH FKORURK\GURFDUERQV H[WUDFWLRQ > @ $V DQ H[WUDFWLRQ SURFHVV VR[KOHW H[WUDFW

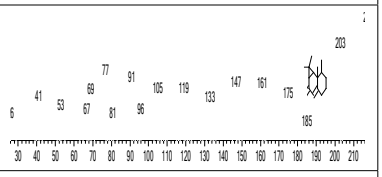
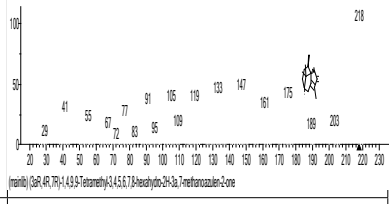
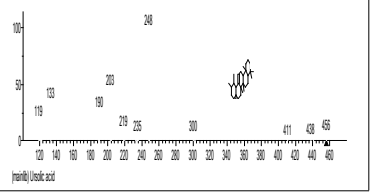
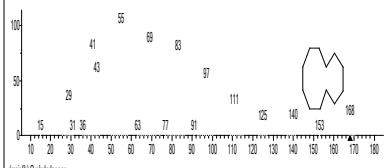
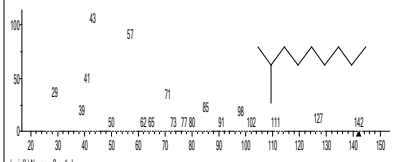
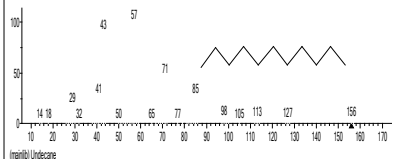
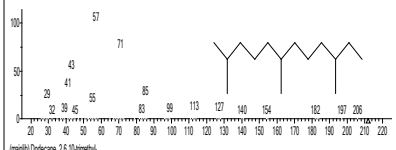
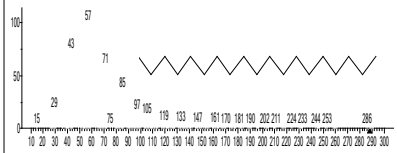
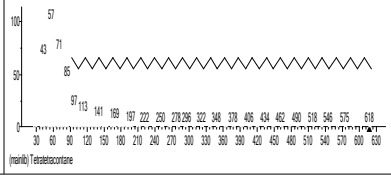
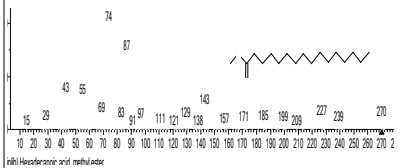
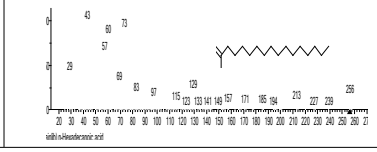
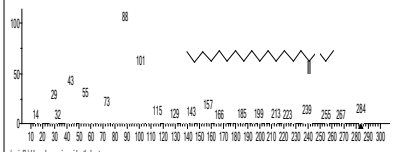
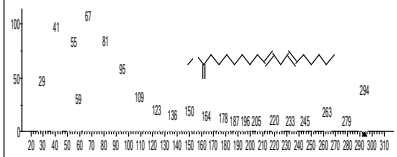
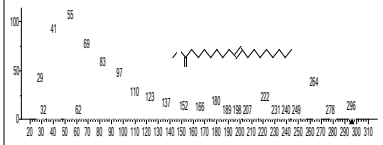
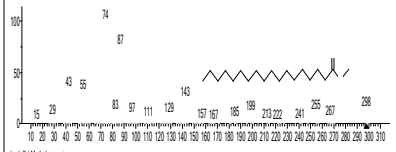
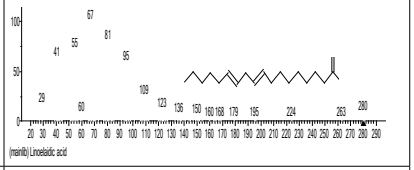
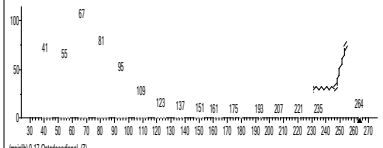
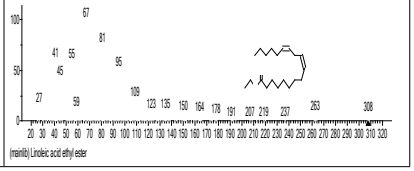
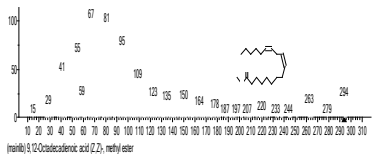
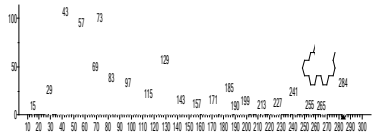
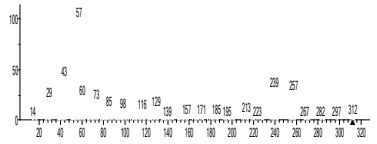
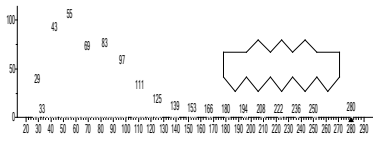
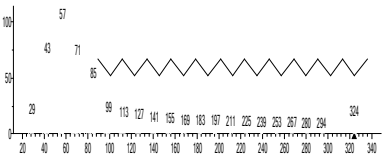
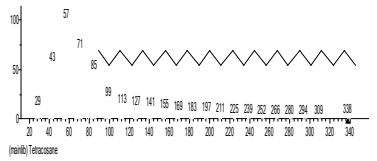
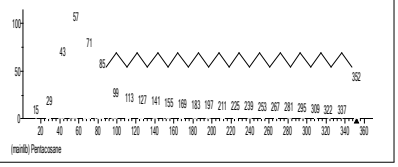
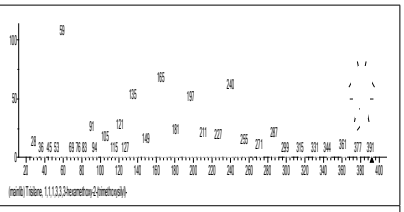
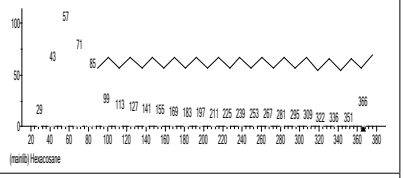
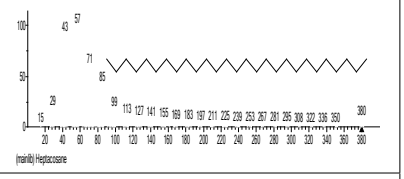
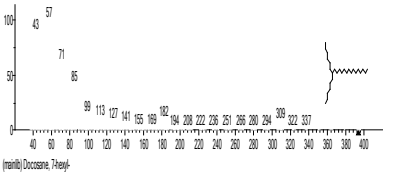
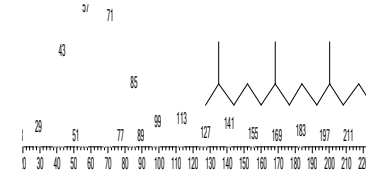
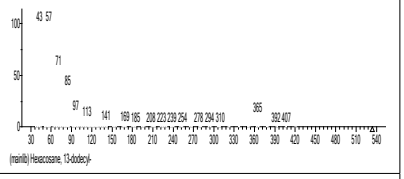
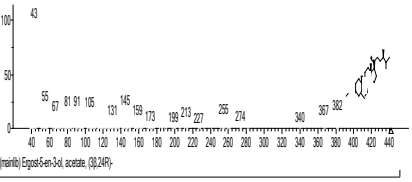
S/N	Compound	Retention Time (min)	Percentage of the total	Molecular formula	Molecular weight	Structure
	+ &\FORSURSD>D@QDSKWKDOHQ RQH			& +		
	D5 5 5 7HWUDPHWK\O KH[DK\GUR + D methane			&8 + 1 3		
3	8V HQ RLF DFLG K\GUR[\ PHWK\O HVWHU EHWD			& + O 3		

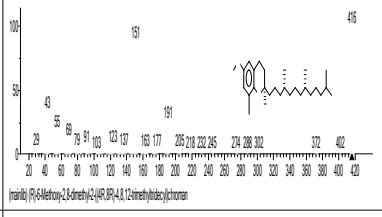
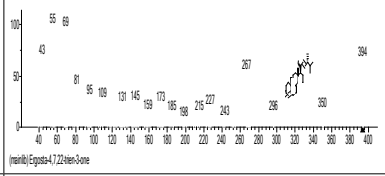
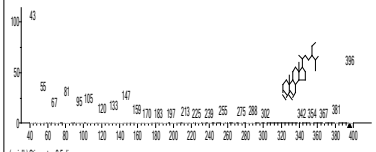
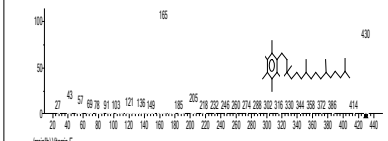
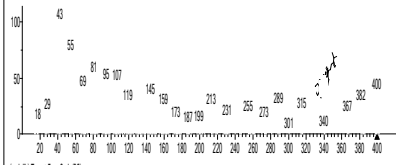
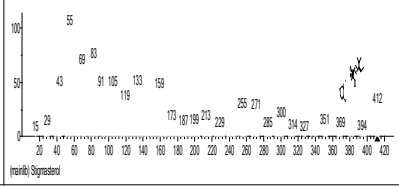
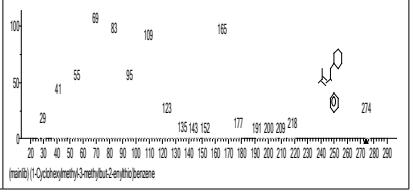
Table 1: Bioactive components of methanol extract of sclerotia of *P. tuberregium*.

S/N	Compound	Retention Time (min)	Percentage of the total	Molecular formula	Molecular weight	Structure
	&\FORGRGHFDQH			& +		
	1RQDQH PHWK\O			& +		
3	8QGHFDQH			& +		
	'RGHFDQH WULPHWK\O			& +		
	2FWDGHFDQH FKORUR			& $+_{37}$ & O		
6	Tetratetracontane			& +		
7	+HSWDPHWK\O ELV WULPHW K\OVLOR[\ WHWUDVLOR[DQH			& + O Si_6		
8	+H[DGHFDQRLF ester	DFLG PHWK\O		& + O		
	Q +H[DGHFDQRLF DFLG			& + O		

	&\FORKHSWDVLOR[DQH tetradeca PHWK\O			& + O_7Si_7		
	+H[DGHFDQRLF DFLG HWK\O ester			& + $_{36}O$		
	2FWDGHFDGLHQRLF acid,methyl HVWHU ((			& + O		
	2FWDGHFHQRLF DFLG PHWK\O HVWHU (			& + $_{36}O$		
	Methyl stearate			& + $_{38}O$		
	+HSWDVLOR[DQH KH[DGHFD PHWK\O			& + O_6Si_7		
	/LQRHODLGLF DFLG			& + O		
	2FWDGHFDGLHQDO =			& + O		
	/LQROHLF DFLG HWK\O HVWHU			& + $_{36}O$		

	2 F W D G H F D G L H Q R L F D F L G = =		3.668	& + O		
	Octadecanoic acid			& $+_{36}$O		
	+ H [D G H F D Q R L F D F L G P H W K \ O propyl ester			& + O		
	& \ F O R H L F R V D Q H			& +		
	& \ F O R Q R Q D V L O R [D Q H P H W K \ O		R F W D G H F D	& + O Si		
	Tricosane			& +		
	Tetracosane			& +		
	, V R S U R S R [\ + H S W D P H W K \ O K \ O V L O R [\ W H W	U D V L O R [D Q H	E L V W U L P H W	& + O_7Si_7		
	3 H Q W D F R V D Q H			& +		

	'LK\GUR[\ benzoic acid, 3TMS derivative			& + O Si$_3$		
	+H[DFRVDQH			& +		
	+HSWDFRVDQH			& +		
	&\FORQRQDVLOR[DQH PHWK\O		RFWDGHFD	& + O Si		
	'RFRVDQH KH[\O			& +		
33	7ULPHWK\OWULGHFDQH			& +		
	&\FORQRQDVLOR[DQH PHWK\O		RFWDGHFD	& + O Si		
	+H[DFRVDQH GRGHF\O			& $_{38}$ + $_{78}$		
36	(UJRVW HQ RO DFHWDWH EHWD 5			& + O		

37	+H[DVLOR[DQH WHWUDGHFDPHWK\O			& + O Si$_6$		
38	5 0HWKR[\ GLPHWK\O 5 5 WULPHWK\OWUL GHF\O FKURPDQ			& + O		
	(UJRVWD WULHQ RQH			& + O		
	6WLJPDVWDQ GLHQH			& +		
	9LWDPLQ (			& + O		
	(UJRVW HQ RO EHWD			& + O		
	Stigmasterol			& + O		
	&\FORKH[\O PHWK\O PHWK\OEXW HQ\OWKLR benzene			& +		

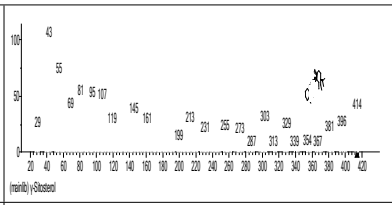
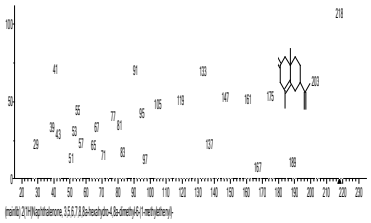
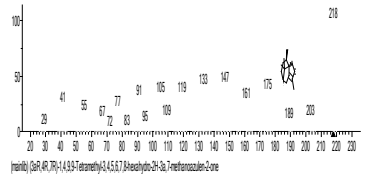
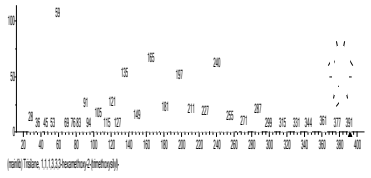
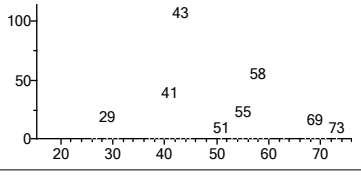
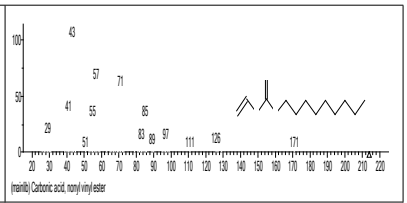
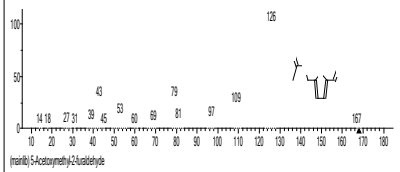
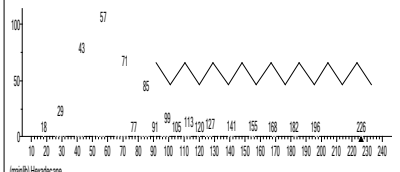
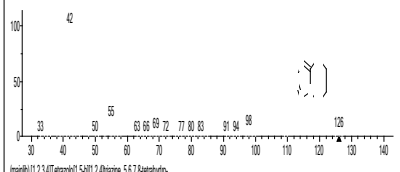
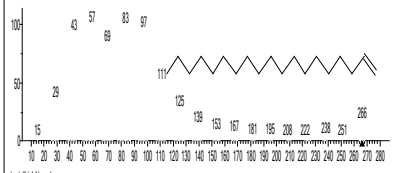
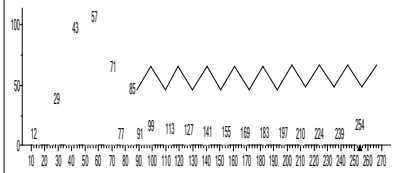
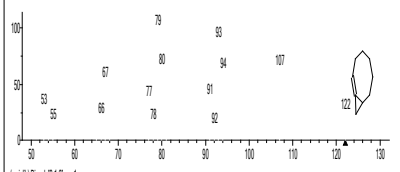
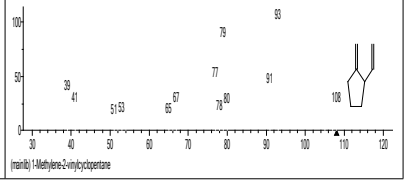
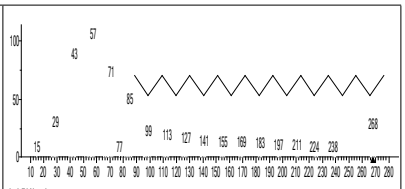
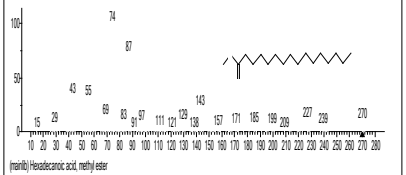
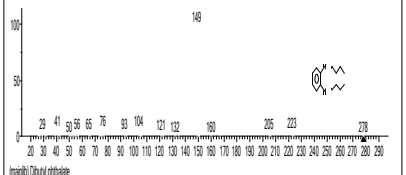
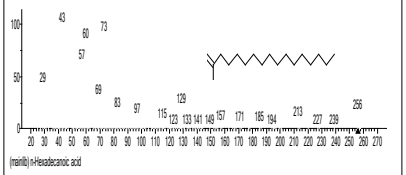
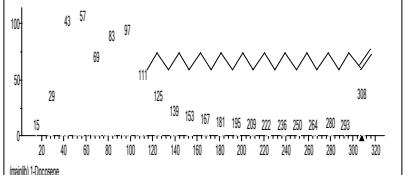
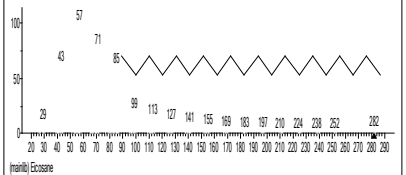
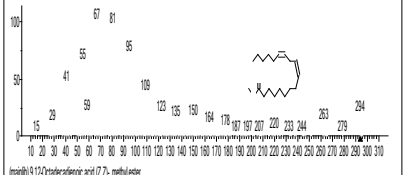
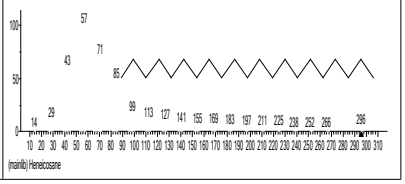
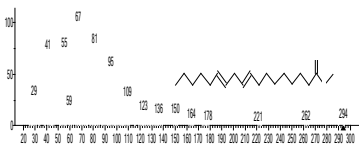
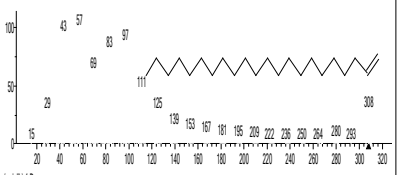
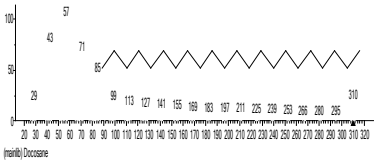
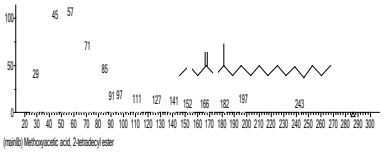
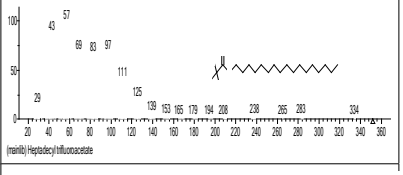
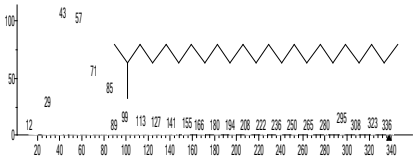
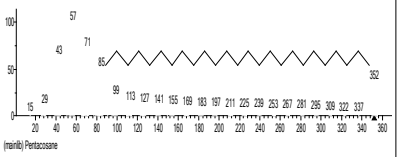
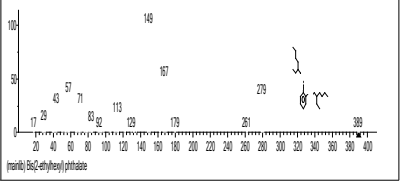
	JDPPD 6LWRVWHURO			& + O		
	+ 1DSKWKDOHQRQH D KH[D K\GUR D GLPHWK\O PHWK\O HWKHQ\O			& + O		
	7KLHQ\O GLK\GUR isoxazo lemethanol			&8 + 12S		
	D5 5 5 PHWK\O KH[DK\GUR + D PHWK DQRD]XOHQ RQH	7HWUD		&8 + 13		
	7HUW %XW\O GLPHWK\OVLO\O GLPHWK\O			& + OSi		

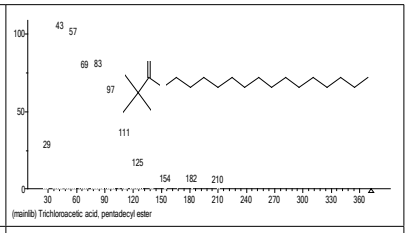
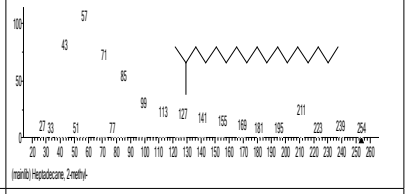
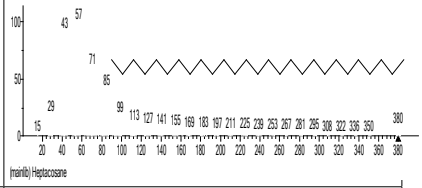
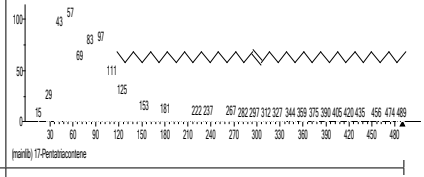
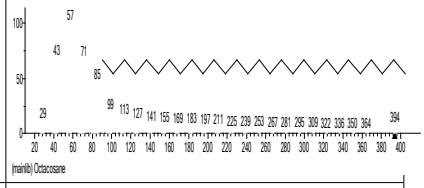
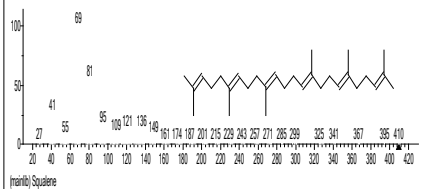
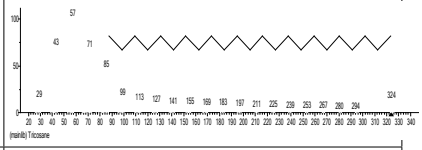
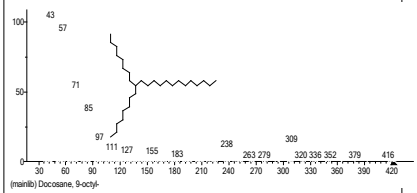
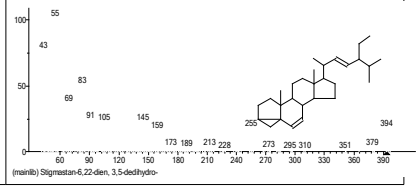
Table 2: Bioactive components of hexane extract of sclerotia of *P. tuberregium*

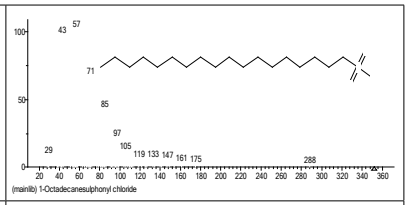
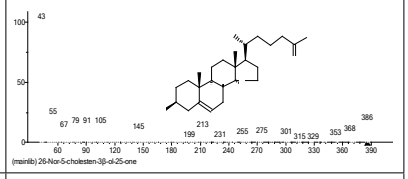
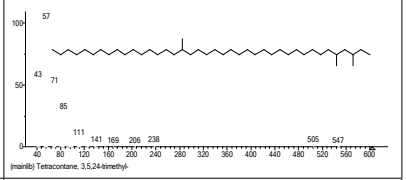
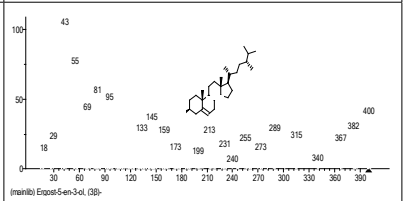
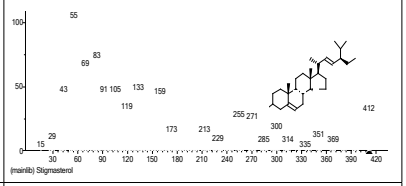
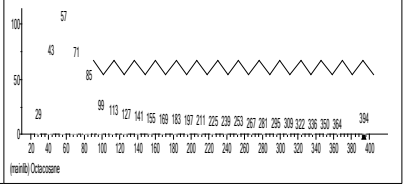
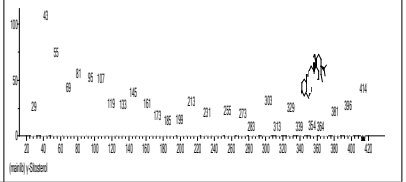
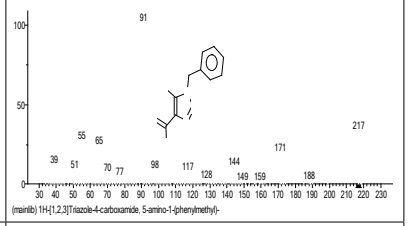
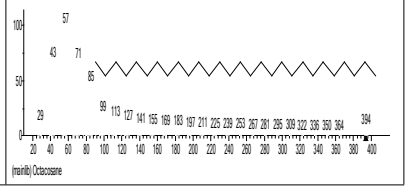
S/N	Compound	Retention Time (min)	Percentage of the total	Molecular formula	Molecular weight	Structure
	&DUERQLF DFLG KH[DGHF\O SURS HQ \O ester			& + O3		

	& D U E R Q L F D F L G nonyl vinyl ester			& + O_3		
3.	$ F H W R [\ P H W K \ O furaldehyde			& $_8$ + $_8$O		
	+ H [D G H F D Q H			& +		
	> @ 7 H W U D] R O R > E @ > @ W U L D] L Q H W H W U D K \ G U R			& $_3$ + $_6$ 1 $_6$		
6.	1 R Q D G H F H Q H			& + $_{38}$		
7.	Octadecane			& + $_{38}$		
8.	% L F \ F O R > H Q H	@ Q R Q		& +		
	0 H W K \ O H Q H vinylcyclo pentane			& $_8$ +		

	1 R Q D G H F D Q H			& +		
	+ H [D G H F D Q R L F D F L G methyl ester			& + O		
	Dibutyl phthalate			& + O		
	Q + H [D G H F D Q R L F acid			& + O		
	' R F R V H Q H			& +		
	Eicosane			& +		
	2 F W D G H F D G L H Q R L F D F L G = = methyl ester			& + O		
	+ H Q H L F R V D Q H			& +		

	2 F W D G H F D G L H Q R L F D F L G = =			& + O		
	' R F R V H Q H			& +		
	Docosane			& +		
	Methoxyacetic D F L G W U L G H F \ O est er			& + O_3		
	+ H S W D G H F \ O W U L À X R U R D F H W D W H			& + F_3O		
	Tricosane, P H W K \ O			& +		
	3 H Q W D F R V D Q H			& +		
	% L V H W K \ O K H [\ O phthalate			& + $_{38}O$		

	Trichloroacetic acid, pentade cyl ester			& + &$_3$O	373.786	
	+ H S W D G H F D Q H P H W K \ O		3.866	& +$_{38}$		
	+ H S W D F R V D Q H			& +		
	3 H Q W D W U L D contene			& +		
	Octacosane			& +		
	Squalene			& +		
	Tricosane			& +		
33	' R F R V D Q H	R F W \ O		& +		
	6 W L J P D V W G L H Q G L K \ G U R	D Q G H		& + O		

	2 F W D G H F D Q H sulphonyl chloride			& + $_{37}$ & OS2		
36	1 R U F K R O H V W H Q E H W D R O R Q H			& + O		
37	Tetracontane, W U L P H W K \ O			& + $_{88}$		
38	(U J R V W H Q R O E H W D			& + O		
	Stigmasterol			& + O		
	Octacosane			& +		
	E H W D 6 L W R V W H U R O			& + O		
	+ 7 U L D] R O H F D U E R [\ O L F D F L G D P L Q R S K H Q \ O P H W K \ O hydrazide			$C_{11}H_{11}N_3O_2$		
	Octacosane			& +		

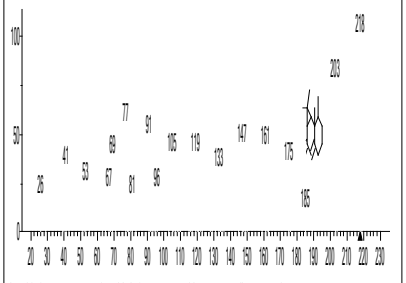
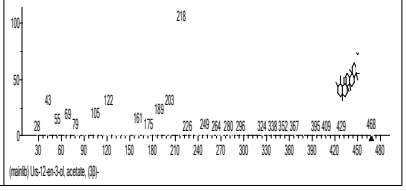
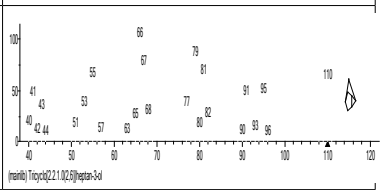
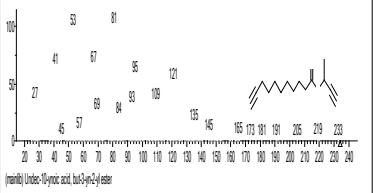
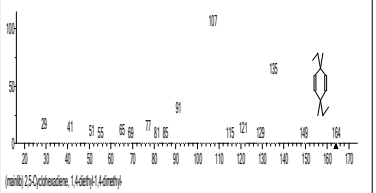
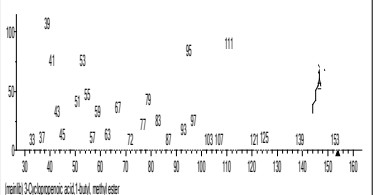
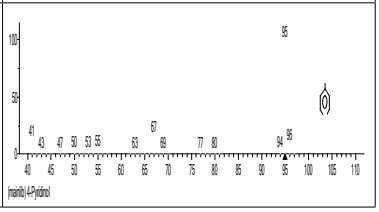
	+ & \ F O R S U R S D > D @ Q D S K W K D O H Q R Q H D D E R F W D K \ G U R D W H W U D P H W K \ O D alpha.,7. alpha.,7a.alpha.,7b. D O S K D			& +		
	8 U V H Q R O D F H W D W H E H W D			& + O		

Table 3: Bioactive components of dichloromethane extract of sclerotia of *P. tuberregium*

S/N	Compound	Retention Time (min)	Percentage of the total	Molecular formula	Molecular weight	Structure
1	Tricyclo[2.2.1.0(2,6)] heptan-3-one, oxime			&, +		
2	Undec-10-ynoic acid, but-3-yn-2-yl ester			& + O		
3	2,5-Cyclohexa diene-1,4-dione, 2,5-dihydroxy-3-methyl-6-(1-methylethyl)-			& + O		
4	3-Cyclopropen oic acid,1butyl, methyl ester			& + O		
5	4-Pyridinol			& + 1 2		

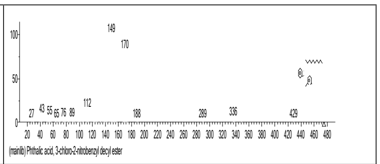
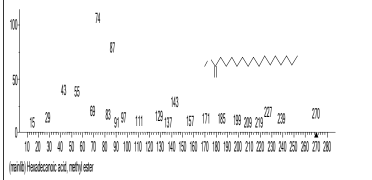
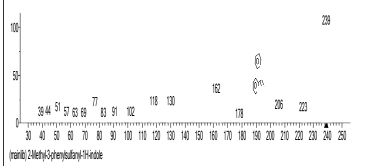
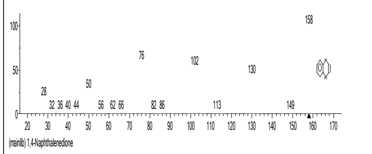
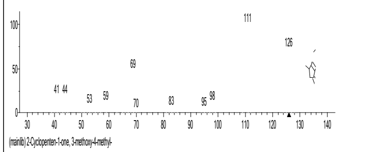
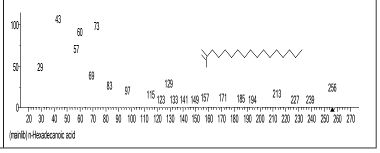
6	Phthalic acid, 3-chlorobenzyl butyl ester					
7	Hexadecanoic acid, methyl ester			& + O		
8	2-Methyl-3-(phenylsulfonyl) methyl-2-cyclopenten-1-one			& 6 + 8 sO		
9	1,4-Naphtha lenedione, 5,8-dihydroxy-2,7-dimethoxy-			& + 6 O		
10	2-Cyclopenten-1-one, 3-methyl-			& + 3 & + O		
11	n-Hexadecanoic acid			& + O		

Table 4: Bioactive components of soxhlet extract of sclerotia of P. tuberregium.

HLWKHU LQVROXEOH RU VSDULQJO\ VROXEOH LQ ZDWHU 7KRXJK VR[UHSHDWHG DSSOLFDWLRQ RI KRW VROYHQW HWKDQRO LQ WKLV FDVH RI WKH VROYHQW LQWR WKH VDPSOH +RZHYHU WKH \LHOG RI WKH H[PD\ EH UHVSRQVLEOH IRU WKH ORZ \LHOG FRPSRXQGV REVHUYHG

CONCLUSION

+H[DQH DQG GLFKORURPHWKDQH H[WUDFWV \LHOGHG PRUH ELRDFWLYH FRPS PHWKDQRO DQG VR[KOHW H[WUDFWV 6HTXHO WR WKHVH ¿QGLQJV KH[DQH D based substrate such as mushrooms and other fungi.

REFERENCES

'DGL ': (PLUH 6$ +DJ (2018) ',PSURYHPHQW RI \LHOG RI ELRDFWLYH F DFWLYLW\ RI PRULQJD VWHQRSHWDOD OHDYHV E\J Food Sci Technol XOW

%RUFKHUV $7 .ULVKQDPXUWK\ $ .HHQ (2008) The immunobiology of mushrooms. 0(([S %LRO 0HG 0D\ZRRG

3. /XOO & :LFKHUV +- (2005) Anti-inflammatory and immunomodulatory metabolites. 0HGLDWRUV ,QÀDPP

2KLUL 5& $PDGL (2018) 3KRWRFKHPLFDO FRQ¿QHPHQW LQ VHFWLRQV WXEHU PXVKURRP Am J Biomed Sci SHFDUS

&KHQ $: +XQJ (2004) 1URGXFWLRQ RI WXEHU OLNH Pleurotus tuberregium RI UPHGLF

6LQJ $JDULFRP\FHWLGHDH *Int J Med Mushrooms*

6. ,VLNKXHPKHQ 62 (2004) %DUEHUUQ'J 3OHXURWXV WXEHUUHJLXP *Mushroom World Web*

7. 2VR 0%$ (1997) 3OHXURWXV WXEHUUHJLXP IURP 1LJHULD *Mycologia*

8. *UHJRUL $ 6YDJHOM 0 (2007) &XOWLYDWLRQ WHFKQLTXHV DQG PHGLFLQDO *Food Tech Biotech*

3DWHO < 1DUDLQ 5 (2012) 0HGLFLQDO SURSHUWLHV RI SOHXURWXV VSH *World J Fungal Plant Biol*

&ORXJK 65 0XOOHQ (2005) +H[DQH/ (QF\FORSHGLD RI WR[LFRORJ\ QG HG

7OLOL $ 6FKLN (2017) The application of dichloromethane and chloroform as reagents in organic synthesis; in: VROYHQWV DV UHDJHQWV LQ RUJDQLF V\QWKHVLV UHDFWLRQV DQG