INTRODUCTION

Exposure to cigarette smoke in utero is associated with numerous adverse pregnancy outcomes ***(Aycicek and Ipek, 2008).*** Active smoking during pregnancy is one of the main causes of preterm labour, premature rupture of memberanes, placental abruption, placenta praevia and delayed intrauterine growth ***(Pichler et al., 2008)***.

Smoking during pregnancy may have even more serious consequences, including spontaneous abortions, growth restrictions, reduced birth weight, increased risks of sudden infant death syndrome (SIDS), as well as long-term behavioural and psychiatric disorders ***(Pattenden et al., 2006).***

Fetal total oxidant status is affected by active or passive exposure to cigarette smoke during pregnancy ***(Aycicek and Ipek, 2008).*** Excessive oxidative stress during pregnancy has been associated with miscarriage and with various pregnancy complications, such as pre-eclampsia, intrauterine growth retardation (IUGR) and diabetes ***(Myatt and Cui, 2009)***.

Increased oxidative stress is associated with an increase in the generation of reactive oxygen species (ROS) and reactive nitrogen species (RNS) which when elevated to a level that

overwhelms tissue antioxidant defense systems ***(Wiesman and Halliwell, 1996)***, induces damaging effects to cellular lipids, proteins and DNA inducing lipid peroxides, protein carbonyls and DNA damage and apoptosis ***(Henson and Johnston, 1987).***

The apoptosis cascade can be triggered by 2 main pathways, via an intrinsic, endogenous system such as the mitochondrial Bax/Bcl-2 or via an extrinsic system Fas and FasL involving trans-membrane receptors of the death receptor family ***(Tews and Goebel, 1997)***. FasL ligand induces apoptosis through cognate interaction with its receptor Fas ***(Hengartner, 2000).***

Bcl_2 and bax are homologous proteins that regulate apoptotic pathways, largely by regulating mitochondrial functions. Bcl_2 inhibits apoptosis, while bax promotes it. The balance between these two proteins will determine the fate of the cells ***(Nagata, 1997; Hockenberyt et al., 1990).***

Oxidative stress has been shown to decrease clonogenic capacity of circulating endothelial progenitor cells (EPCs) ***(Ingram et al., 2006),*** which play a pivotal role in the maintenance of endothelium integrity, repair after injury and postnatal neovascularization ***(Hirschi et al., 2008).***

EPCs circulate in peripheral blood and home to sites of neovascularization, including ischemic tissues and tumor microenvironments ***(Sabatier et al., 2009).*** Among different subtypes of EPCs, both the role of angiogenic mononuclear cells (triple-positive CD34+CD133+VEGFR-2+ ***(Zampetaki et al., 2009).*** It is suggested that EPCs play an important role during neonatal development ***(Borghesi et al., 2010)*** and that their concentrations may affect birth weight ***(Aroviita et al., 2010).***

Cord blood is neonatal peripheral blood, where at birth, the cytokine responses of cord blood mononuclear cells to innate/adaptive stimuli tend to reflect the maternal/neonatal environment ***(Gold et al., 2009).*** It is suggested that the cord blood volume, total number of nucleated cells, and EPCs cells are influenced by maternal/neonatal obstetric factors. Significantly positive correlations were observed among the neonatal weight, placental weight, total number of nucleated cells, and EPCs cells ***(Omori et al., 2010).*** Accordingly this study is conducted to measure the impact of maternal smoking induced oxidative stress on EPCs.

AIM OF THE WORK

To evaluate the effect of active and passive smoking by the mother throughout pregnancy on oxidative stress induced apoptosis and EPCs in umbilical cord blood collected at term.

SMOKING

Introduction:

Smoking is a major world wide public health problem. The period from early teens to early twenties is a critical period in which the majority of individuals begin to develop the social pattern that will cause them to start smoking or to become smoker later on their lives ***(Azab et al., 1995).***

Tobacco use is one of the leading preventable causes of premature death, disease, and disability around the world. Nearly five million deaths every year can be attributed to tobacco use, and many more suffer from smoking-related morbidity, while the number of fatalities is expected to more than double by the year 2020, if the current epidemic continues, more than 70 % of these deaths are expected to occur in developing countries ***(Ljaljvi et al., 2008).***

The median age of smoking initiation in many countries is below the age of 15, and smoking prevalence rates are rising among children and adolescents. If we do not deal with this problem effectively, tobacco use will result in the deaths of 250 million children and adolescents alive today, mostly in developing countries ***(Dous, 2003).***

Tobacco consumption has scored many records and it is still progressing. Egypt is the highest consumer of tobacco in the Middle East and North Africa (MENA) region ***(Islam and Johnson, 2005).***

In Egypt, the number of smokers is steadily increasing by 8% every year, with a declining age of initiation ***(Islam and Johnson, 2005).***

Epidemiology of smoking:

Prevelance of smoking world wide

Prevalence of tobacco consumption is reported by the World Health Organization (WHO), which focuses on smoking (not smokeless chewing tobacco) due to reported data limitations. Smoking has therefore been studied more extensively than any other form of consumption ***(World Health Organization, 2009).***

Smoking is generally five times higher among men than women ***(Guindon and Boisclair, 2003).*** However the gender gap declines with younger age ***(Surgeon General's Report, 2001).*** In developed countries smoking rates for men have peaked and have begun to decline, however for women they continue to climb ***(Richard et al., 2006).***

Smoking prevalence has changed little since the mid-1990s (until which time it declined in English-speaking countries, which have all implemented tobacco control) ***(Robert and Shiffman, 2007).***

As of 2002, about twenty percent of young teens (13–15) smoke worldwide. 80,000 to 100,000 children begin smoking every day. Half of those who begin smoking in adolescent years are projected to go on to smoke for 15 to 20 years ***(World Health Organization, 2009).***

The World Health Organization (WHO) states that "Much of the disease burden and premature mortality attributable to tobacco use disproportionately affect the poor". Of the 1.22 billion smokers, 1 billion of them live in developing or transitional economies. Rates of smoking have leveled off or declined in the developed world ***(Centers for Disease Control and Prevention, 2009).*** In the developing world, tobacco consumption is rising by 3.4% per year as of 2002 ***(World Health Organization, 2009).***

The WHO in 2004 projected 58.8 million deaths to occur globally from which 5.4 million are tobacco-attributed ***(Global Burden of Disease, (2004).*** As of 2002, 70% of the deaths are in developing countries ***(World Health Organization, 2009).***

It is predicted that 1.5 to 1.9 billion people will be smokers in 2025 ***(Guindon et al., 2009).***

Table (1): Smoking prevalence by gender:

	Percent smoking	
Region	**Men**	**Women**
Africa	29	4
United States	35	22
Eastern Mediterranean	35	4
Europe	46	26
Southeast Asia	44	4
Western Pacific	60	8

(World Health Organization, 2000)

Epidemiology of smoking in Egypt:

Smoking prevalence among adult men in Egypt is 43.4 %, and it is 4.7% among women ***(Islam and Johnson, 2005).*** Others estimated it to be 47% among males over 15 years old ***(Mohamed et al., 2006).***

Women from low socioeconomic classes had more intense and longer exposure than women from high socioeconomic classes mainly because of work exposure ***(Curtin et al., 1998).***

Cigarettes consumption:

Tobacco consumption has scored many records and it is still progressing. Egypt is the highest consumer of tobacco in the Middle East and North Africa (MENA) region ***(Islam and Johnson, 2005).*** Egypt has the highest rate of tobacco consumption in the Arab world ***(World Health Organization, 2000).***

Smoking in Egypt is prevalent, as 19 billion cigarettes are smoked annually in the country, making it the largest market in the Arab world ***(Knell, 2010).***

In the past few years, smoking in Egypt has reached an all time high with an estimated twenty percent, ten million people, regularly using tobacco products ***(Egypt Global Adult Tobacco Survey, 2012).***

Egypt is ranked as one of the top ten per capita consumers of tobacco by the World Lung Foundation. Of this twenty percent of the population estimated to use tobacco products ninety-five percent were daily smokers. Sixteen percent smoke only cigarettes, 3.3% smoke shisha water-pipes, and 2.6% use smokeless tobacco products. While consumption of tobacco in adults is in decline or stagnant in many countries, in Egypt the number of adult cigarette smokers is increasing at a rate of four to five percent per year ***(Ahmed, 1999)***.

Types of smoking:

Active smoker

Active smoker who smoked one or more cigarettes per day ***(Anastasia et al., 2009).***

Passive smoker

Passive smoking is defined as an involuntary exposure to environmental tobacco smoke ***(Krisela et al., 2006).***

Passive smoking has been shown to adversely affect the health of infants and children ***(Jennifer et al., 1996).***

Passive smoking results in inhalation of nicotine by children in amounts equivalent to their actively smoking 60 to 150 cigarettes yearly ***(Jennifer et al., 1996).***

Methods of tobacco smoking:

SNUFF:

Snuff consists of ground dried tobacco leaves of the tobacco species Nicotina tobacum and frequently contains additives ***(Krisela et al., 2006).***

Prolonged use cause atrophy of the nasal mucous membrane with replacement of ciliated columnar by squamous epithelium ***(Laurence and Bennett, 1992).***

Cigars and pipes:

They have alkaline PH (8.5) and nicotine is relatively unionized and lipid soluble so that is readily absorbed in the mouth. Cigar and pipes smoker thus obtain nicotine without inhaling (they also have a slower death rate from lung cancer) ***(Laurence and Bennett, 1992).***

Cigarettes:

A **cigarette** (French "small cigar", from *cigar* + *-ette*) is a product consumed through smoking and manufactured out of cured and finely cut tobacco leaves and reconstituted tobacco, often combined with other additives, then rolled or stuffed into a paper-wrapped cylinder (generally less than 120 mm in length and 10 mm in diameter). The cigarette is ignited at one end and allowed to smolder for the purpose of inhalation of its smoke from the other (usually filtered) end, which is inserted in the mouth. They are sometimes smoked with a cigarette holder. The term cigarette, as commonly used, refers to a tobacco cigarette but can apply to similar devices containing other herbs, such as cannabis ***(Wigand, 2006).***

They are acidic (ph 5.3) and nicotine is ionized and insoluble in lipids most cigarettes contain 6 to 8 mg of nicotine ***(Herfindal and Gourley, 2000).***

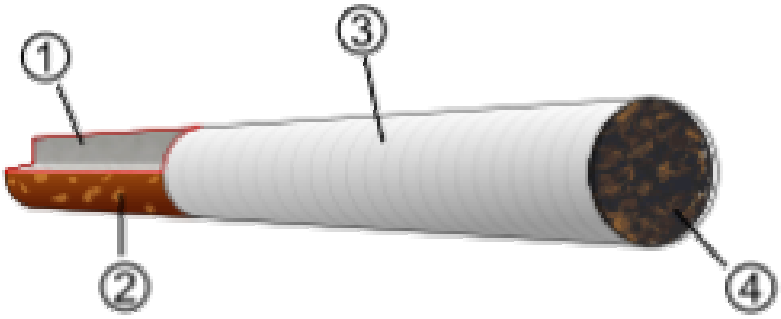

Fig. (1): Diagram of a cigarette. (1) Filter made of 95% cellulose acetate. **(2)** Tipping paper to cover the filter. **(3)** Rolling paper to cover the tobacco. **(4)** Tobacco blend ***(Wigand, 2006).***

Chewing tobacco:

Nicotine in chewing tobacco is more slowly absorbed than inhaled nicotine, so it has a longer duration of effect ***(Goodman and Gilman, 1996).***

Narghil (hubble-bubble) smoking:

The waterpipe (also known as gouza, narghile, hubble-bubble, hookah or shisha, depending on the local tradition) has been used for smoking tobacco for centuries in the Eastern Mediterranean Region.

There are three distinct types of water pipe. The gouza is the oldest form of water pipe. It has a small water container (about 200–500 ml**)** that is made of metal; coconut shell was formerly used. The bouri has a water container made of brass (about 200–

500 ml). The authentic shisha is a larger **(**about 1000–2000 ml) and more decorated form of water pipe, usually with a glass water contain It's reported that 11% of Egyptian men and women smoke shisha ***(Lancaster, 1996)*** **(**Fig. 2).

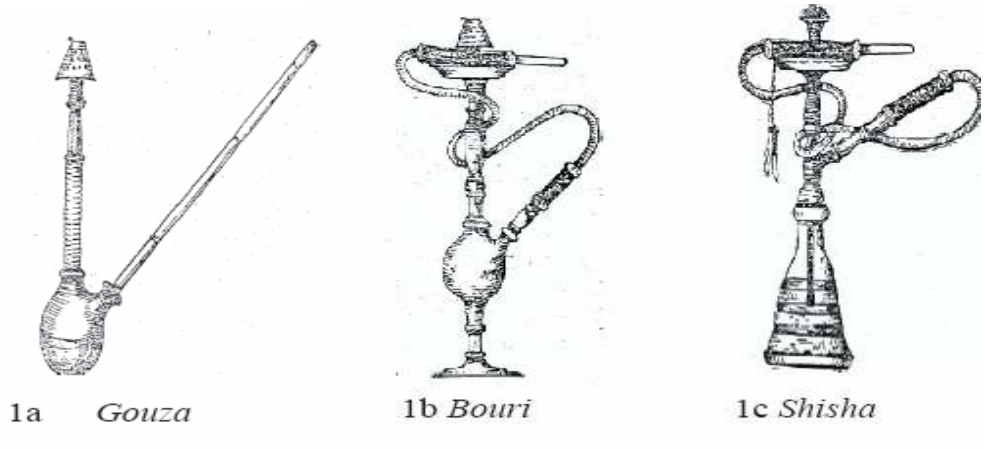

Fig. (2): Different types of waterpipe ***(Lancaster, 1996)***

Earlier studies have reported an association between shisha smoking and oral cancer ***(Gupta et al., 1996)*** and decreased fertility ***(Inhorn and Buss, 1994),*** these effects may be due to higher carboxyhemoglobin **(COHb)** compared to cigarette smoking ***(Zahran and Al Fayez, 1985).***

Chemical composition of tobacco:

Tobacco smoke components are distributed between the particulate phase and the vapour or gas phase of the smoke aerosol.

a) Particulate phase and condensable substances:

The portion of smoke which is retained on a Cambridge filter pad at room temperature.Substances with MW > 200 g/mol ***(Institute for Health and Consumer Protection, 2007).***

b) The Gaseous Phase (not condensable):

For the purposes of a generally accepted definition, the portion of smoke that passes through a Cambridge filter pad at room temperature is called the vapour phase. Substances with MW < 60 g/mol ***(Institute for Health and Consumer Protection, 2007).***

Main stream and side stream smoke:

Main stream smoke:

Mainstream smoke was defined as the smoke that is inhaled by the smoker ***(Michael et al., 2005)*** emerges into the environment after having been drawn through the cigarettes filtered by the smokers on lungs and then exhaled ***(Brunneman et al., 1978).***

Side stream smoke:

Arises from the burning end of the cigarette and enters directly into the environment ***(Brunneman et al., 1978)***, while patients with side stream smoke exposure were defined as those who self-identifying that they live with a partner who regularly smokes ***(Michael et al., 2005).***

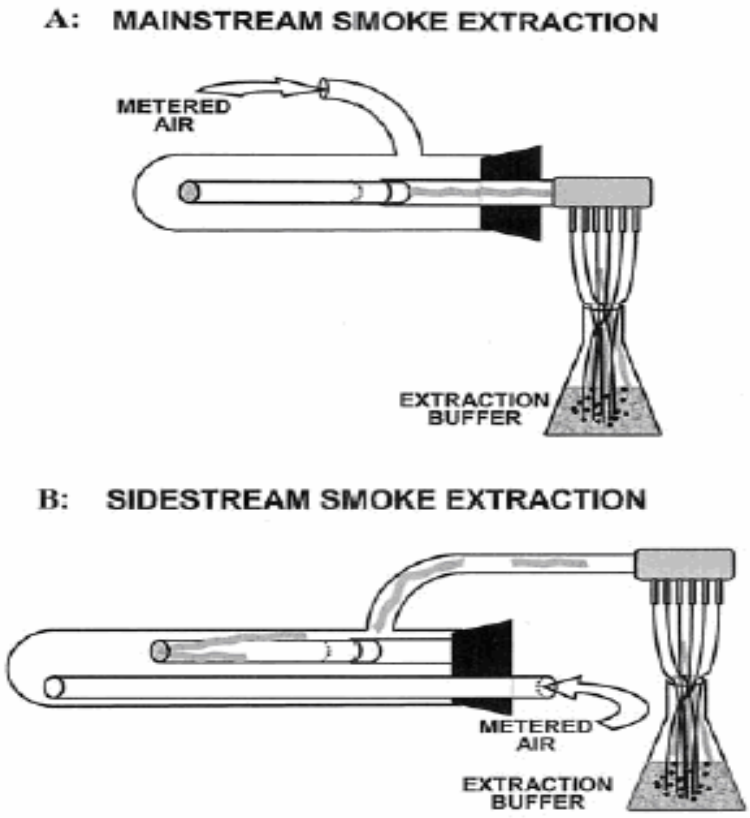

Fig. (3): Schematic of smoke extraction methods. **(A)** In mainstream extraction, all smoke passes through cigarette filter. **(B)** In sidestream extraction, no smoke passes through filter ***(Michael et al., 2005).***