Nicotine is the most important pharmacologically active compound in all forms of tobacco smoke because of its interaction with cholinergic receptors. In the current issue of the Journal, a research article by Lee et al. (1) and a brief communication by Falvella et al. (2) use different approaches to contribute to a better understanding of the roles of nicotine receptors and the biological basis that underlies the environmental and genetic components of smoking-related cancer risk.

Two types of acetylcholine receptors (AChRs) exist: the nicotinic AChRs (nAChRs) and muscarinic AChRs (mAChRs). The nAChRs are ligand-gated ion channels and form pentameric complexes that consist of four related but genetically and immunologically distinct subunits: α, β, γ, and δ (3). The α9-nAChR, a known homopentamer that plays a central role in coordinating keratinocyte adhesion and motility during wound healing (4), is encoded by a gene (CHRNA9) on human chromosome 4p (5). Lee et al. (1) showed that α9-nAChRs were ubiquitously expressed in many epithelial and cancer cell lines from lung and breast and that most of the same cell lines also expressed α5- and α10-nAChRs. The α9-nAChRs were also present in primary tumors and non-malignant breast tissue obtained from patients; however, cancers had increased α9-nAChR expression compared with the surrounding normal tissues. Confocal microscopy revealed membranous colocalization of α9-nAChR protein with caveolin in MCF-7 breast cancer cells, whereas the immunohistochemical localization in tissue samples was somewhat equivocal. The expression data set a foundation for the functional experiments that followed.

Exposure to nicotine increases the expression of α9-nAChR in breast cancer cells (6,7). Nicotine itself is not a carcinogen, but it promotes the growth of cancer cells and the proliferation of endothelial cells (8,9). In addition, nicotine and tobacco-specific nitrosamine 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone (NNK) bind to nAChRs with higher affinity than their natural ligand, acetylcholine. In the current article (1), Lee et al. used MDA-MB-231 breast cancer cells in which α9-nAChR expression had been silenced to show that lowering expression reduces proliferation and tumorigenic potential in both in vitro and in vivo assays. Cells with inducible α9-nAChR gene expression were also generated from normal breast epithelial cells (MCF-10A) that were transformed by nicotine or NNK treatments. They were assayed to show that increased expression of the α9-nAChR in vitro enhances proliferation and colony formation. Likewise, exposure to nicotine among mice injected subcutaneously with nicotine-transformed MCF-10A cells that inducibly express increased levels of α9-nAChRs further enhanced the volume of tumor xenografts. This raises an important question: Were the “transformed” nicotine- or NNK-treated MCF-10A cells now cancer cells or still precancerous, as suggested by the authors? The current paper by Lee et al. does not further address the molecular mechanism, but other groups have reported that nicotine decreases the cytotoxicity of doxorubicin, promotes migration via a signaling cascade involving protein kinase C and cdc42, and induces proliferation, invasion, and epithelial–mesenchymal transition in breast cancer cells (8,10,11). Lee et al. have provided evidence that the α9-nAChR may play a major role in breast carcinogenesis, just as the α7-nAChR is often associated with lung cancer (12). This further supports epidemiological studies that have revealed an association with breast cancer and exposure to cigarette smoke (13).

Binding of nicotine to nAChRs also forms the basis of the molecular pharmacology that leads to smoking addiction. The findings from several genetic studies have suggested that variants on chromosome 15q24–25, which includes the α5-α3-β4 nAChR cluster, contribute to nicotine addiction and to risk of lung cancer and chronic obstructive pulmonary disease. Although multiple genes and polymorphisms are likely to be involved, it is not easy to reveal causal relationships (14–17). In a recent article, Impuro et al. (18) revealed a specific role for achaete–scute complex homolog 1 (ASCL1), a neural transcription factor implicated in the development of neural tissue and small cell lung carcinoma, a cancer which is strongly associated with smoking, in regulating the expression of the nAChR cluster (18). Falvella et al. (17) showed that expression of the CHRNA5 gene, which encodes the α5-nAChR, was increased in lung adenocarcinoma tissues and that the D398N polymorphism in CHRNA5 was associated with lung cancer risk. In the current issue of the Journal (2), Falvella et al. associated haplotypes from the 5′ promoter region with CHRNA5 transcript levels in primary lung tissues. In vitro, polymorphisms in the CHRNA5 promoter strongly modulated its activity as indicated by luciferase assays. The association of these functional polymorphisms with lung cancer risk may help us in the future to understand the mechanisms associated with the risk of this disease.

The fact that Lee et al. (1) were able to show that nicotine enhances the growth of breast cancer cells via α9-nAChRs suggests not only that smoking could be causally related to breast carcinogenesis but also that nicotine could directly contribute to the molecular mechanism of carcinogenesis in addition to indirectly contributing by promoting addiction to smoking. In addition, the studies by Falvella et al. (19) indicate that some of the polymorphisms associated with nicotine addiction and susceptibility may increase the expression of α5-nAChRs in parenchymal tissue from lung cancer patients. Although we do not know what regulates the
assembly of nAChR subunits, they regulate Ca\(^{2+}\) influx and intracellular Ca\(^{2+}\) concentration to different degrees and work through receptor-specific downstream signaling pathways (3). Moreover, the receptor complex may change as a function of the cell's differentiation status (20). In the brain, cholinergic receptors bind to acetylcholine or possibly to nicotine to mediate chemical transmission in a highly interactive process (21). Cholinergic signaling in non-neuronal cells is comparable to cholinergic neurotransmission (22). Dysfunction of this system is involved in the pathogenesis of disease ranging from nicotine addiction to cancer (21,23). Better understanding the molecular mechanisms of the cholinergic pathways will lead to more opportunities for intervention and prevention of tobacco toxicity.

References


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