Heart–Lung Interaction via Infection

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Abstract

Lung and cardiovascular disease are increasingly recognized to occur in the same patient populations. Infections, either through stimulation of inflammation or through direct infection, can lead to end-organ damage and have been postulated as a potential link between lung and cardiovascular diseases. Mechanisms by which infections may link lung and cardiac diseases include effects of systemic infections, microbial translocation of pathogens from the gastrointestinal tract or other sites, damaging effects of metabolic products, or influences of smoking on the microbiome.

Other mechanisms, such as alterations in the local microbiome, environmental exposures, or immune regulation by microbial communities, may be important. These relationships are likely quite complex, with multiple routes between infection and disease possible. A better understanding of the links of infection to lung and heart disease can improve our understanding of the pathogenesis of these disorders and uncover novel therapeutic approaches.

Keywords: microbial translocation; metabolomics; pulmonary function; cardiovascular; HIV

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Lung and cardiovascular diseases are increasingly recognized as occurring in concert in many individuals. Whether these diseases develop as a result of unique mechanisms or shared pathways remains uncertain, but growing evidence indicates that they may share common origins. Although environmental exposures such as cigarette smoking may lead to lung and cardiovascular disease (CVD), the relationship between diseases in these two organ systems seems to have a component independent of smoking and likely results from other shared host or environmental causes. Infections, including systemic pathogens and the microbiota of various body niches, may contribute to the development of lung disease and CVD directly or through stimulation of inflammation. Understanding the mechanisms of infection in lung and CVD could identify novel treatment modalities for these diseases, such as antibiotics or probiotics. This review discusses the links of

lung disease and CVD, reviews the potential roles of infections, and provides examples to support various pathogenic mechanisms.

Clinical Links of Lung and Cardiovascular Diseases

Chronic obstructive pulmonary disease (COPD) frequently coexists with cardiac diseases, including atherosclerosis and pulmonary hypertension. The coexistence of COPD and CVD is acknowledged as an important clinical syndrome that is likely stimulated by common inflammatory pathways and is associated with poor outcomes, including increased use of health resources and mortality (1–5). COPD is associated with adverse cardiovascular effects, including increases in endothelial dysfunction, arterial stiffness, and atherogenesis, which are independent of the effects of cigarette smoking (1, 6–8). FEV₁ is an important independent predictor of

CVD even in nonsmokers, and smokers with COPD are more likely to have CVD than smokers without COPD (9). A metaanalysis of large studies of CVD found an increased risk of cardiovascular mortality in smokers and nonsmokers in the lowest quintile of FEV₁ (10). COPD is also an independent risk factor for increased carotid intima medial thickness (5, 6). Decreases in endothelial reactivity as measured by flow-mediated dilation have been correlated with decreases in FEV_{1%} predicted and FEV₁/FVC and increases in CT-documented emphysema independent of smoking history (8, 11). Pulmonary hypertension is increased in the COPD population as well (12, 13). Increases in systemic inflammatory markers, such as high-sensitivity C-reactive protein, IL-8, and IL-6, and increases in sputum neutrophils in COPD are associated with reduced lung function (14, 15). These findings suggest that chronic inflammation may be the common pathway to end-organ

damage in these conditions directly by release of damaging cell products or via effects on vascular cell activation and apoptosis, increased thrombogenicity, or oxidative stress. Although this inflammation may result from multiple causes, infections are likely candidates to initiate or perpetuate such inflammation.

Hypothesized Mechanisms of Heart–Lung Interactions via Infection

There are a multitude of potential pathways that could link lung and heart disease via infections (Figure 1). For example, systemic infections could stimulate inflammation and immune activation, thus worsening lung and heart disease. Alternatively, localized lung infections could result in or worsen CVD via stimulation of local inflammation that “spills over” and becomes systemic. Microbial translocation of bacteria or other organisms from the gastrointestinal (GI) tract or other sites could travel to the lung or the vasculature and directly infect tissues. Translocated microorganisms could also stimulate local and systemic inflammation, resulting in tissue damage. Metabolic products of

microbes or the resulting inflammation could alter the course of lung and CV diseases. Metabolomic effects could result from translocated bacteria or from systemic products of microbial communities in various body habitats. Alterations in lung and GI microbial communities resulting from environmental exposures, including cigarette smoking and pollution, could also influence disease. The composition of the GI microbiota shapes development of the immune system and could influence lung and CVD via this route. A detailed discussion of the literature supporting the various aspects of all possible routes from infection to lung and heart disease is beyond the scope of this review, but we highlight specific examples of possible links between lung and heart disease via infection (Table 1).

Effects of Systemic Infection

HIV as a Representative Infection

One potential way in which infection could link lung and heart disease is via systemic infection. Chronic systemic infections could promote disease via activation of systemic inflammation that leads to end-organ damage. An example of one such infection

is HIV. Lung and cardiac diseases, including CVD and pulmonary hypertension, have been reported to be accelerated in individuals with HIV and seem to occur together (16–20). For example, in a cohort of HIV-infected outpatients, we found that pulmonary function is significantly associated with multiple aspects of cardiovascular disease, including coronary artery calcification and echocardiographic evidence of pulmonary hypertension (17; Morris, unpublished data). HIV-infected participants with elevated pulmonary artery pressures on echocardiography (defined as tricuspid regurgitant jet velocity [TRV] ≥ 3.0 m/s) had significantly worse airflow (17). This relationship with abnormal lung function was similar with pulmonary artery systolic pressure (PASP). Studies have shown that particular mutations in the *nef* allele of HIV are associated with pulmonary hypertension and that *nef* can be found in endothelial cells from HIV-infected patients with pulmonary hypertension and in a nonhuman primate model (21, 22), suggesting that the direct effects of HIV may contribute to pulmonary hypertension.

Relationship of Inflammation

It is possible that chronic immune activation and inflammation from the virus or its coinfections could cause lung and cardiac disease, but there are complex mechanisms involved, including effects of immunosuppression, coinfections, antimicrobial prophylaxis, and antiretroviral therapy (ART) for HIV. Lung and CVD in HIV appear related to local and systemic inflammation, although a direct role for HIV in stimulating this inflammation is not yet proven. The inflammation may be exaggerated by smoking, but does not depend on smoke exposure. In one study of HIV-infected individuals, lower DL_{CO} was associated with greater sputum neutrophils in smokers and nonsmokers (23). We also examined the relationship of pulmonary artery pressure measurements to pulmonary and systemic inflammation in the cohort of HIV-infected individuals described above (17). Percent sputum neutrophils tended to increase with increasing TRV, and increasing sputum IL-8 was significantly associated with higher PASP and TRV. Plasma IL-8 was also significantly associated with increasing PASP and TRV. These studies suggest that HIV may be a systemic infection linked to lung and heart disease via inflammation.

Figure 1. Possible pathways linking lung and heart diseases via infection, including direct effects of lung infection, stimulation of systemic inflammation, translocation of gastrointestinal microbes, and effects of metabolomics pathways.

Table 1. Potential mechanisms linking lung and heart disease via infection

Potential Mechanism	Examples of Related References
Direct effects of pathogens (local, systemic, or translocated)	16–23, 25, 26, 29, 32–34, 39, 40
Stimulation of systemic inflammation	7, 23, 25, 26, 29, 39
Alterations in microbiota of lung, gastrointestinal tract	32, 35, 36, 41, 42
Metabolic products of microbiota or inflammation	41–44
Alterations of microbiota from cigarette smoking	45–48

The Role of Microbial Translocation

Microbial translocation (MT) is commonly defined as transmucosal passage of microorganisms and/or microbial products across a mucosal surface. Although most commonly studied in the GI tract, translocation could occur via other mucosal surfaces, such as the mouth, lung, or urogenital tract. Microbial translocation could lead to heart and lung disease by direct infection of these organs or via stimulation of inflammation.

Bacteria and the GI Tract

Translocation of bacteria is assessed by measurement of LPS (a component of the outer bacterial membrane that is strongly immunogenic) levels, detection of bacterial 16S ribosomal DNA (a ubiquitous gene in bacteria) in peripheral blood, or measurement of soluble CD14 (a receptor for LPS). Bacterial translocation from the GI tract to the peripheral circulation has been extensively studied in HIV infection, but is also common in liver disease and other conditions (24–27). Translocation occurs early in HIV infection due to depletion of gut CD4⁺ lymphocytes and is not completely eradicated by ART, particularly in patients with a suboptimal CD4 cell count or HIV viral level responses (25, 28). Persistent systemic exposure to bacterial antigens from MT triggers immune activation and inflammation in HIV-infected adults (25, 26). MT is associated with T-cell activation, which predicts systemic inflammation, mortality, HIV progression, and progression of comorbidities such as cardiovascular disease (25, 26, 29–31).

Although direct evidence of GI translocation in HIV resulting in lung infection is lacking, a recent study from the

Lung HIV Microbiome Program found evidence of *Tropheryma whippelii* in bronchoalveolar lavage (BAL) from an HIV-infected ART-naïve individual (32). *T. whippelii* was also found to be much more prevalent in BAL samples from HIV-infected individuals than from HIV-uninfected individuals in a multicenter cohort from this group (32). *T. whippelii* is the agent of Whipple's disease, a predominantly GI condition that is rarely seen in the lung. Although stool studies were not performed in the larger cohort and the stool sample for the initial *T. whippelii*-infected individual did not contain *T. whippelii* DNA, it is possible that the organism previously had translocated from the GI tract. Future studies are needed to determine the origin of *T. whippelii* in HIV and its relationship to lung disease.

The potential for GI bacteria to infect the lung via translocation has been confirmed by animal models showing that inoculation of the gut with pathogenic bacteria can result in pulmonary infection with the same organisms. In a study of newborn rabbits, animals fed with traditional formula and gavaged with *Enterobacter cloacae* had *E. cloacae* in blood, lung, and other tissues by traditional culture (33). Animals also fed a probiotic had decreased detection of *E. cloacae*. These findings illustrate the ability of GI bacteria to translocate to the lung and suggest the potential for therapy of lung infections via modulation of GI microbiota.

Fungi as Possible Agents of Microbial Translocation

Although bacteria translocation is common in HIV, it is also possible that fungal translocation from mucosal surfaces of the GI, respiratory, and genitourinary tracts or from diseased skin could occur. HIV-infected individuals are at increased risk of

oral and esophageal candidiasis and other fungal diseases, and *Candida* is a normal colonizer of mucosal surfaces. We have also found that the fungus *Pneumocystis* is a frequent respiratory colonizer in HIV-infected individuals, particularly in those with COPD (34–36). (1→3)- β -D-glucan (BG) is a component of fungal cell walls that is shed into the blood and can be detected in the serum of patients with fungal pneumonia (37, 38). We have recently found detectable BG levels in HIV-infected adult outpatients without pneumonia, and BG levels correlate strongly with airway obstruction, DL_{CO}, echocardiographic pulmonary artery pressures, higher inflammatory cytokines, increased sputum neutrophils, and activated peripheral lymphocytes (39). These results indicate that fungal translocation is an unrecognized contributor to inflammation and chronic complications of HIV.

Viruses as Possible Agents of Microbial Translocation

Viral translocation may occur, but this process has not been extensively studied. One study examined intratracheal inoculation of Apo E-deficient mice with influenza (40). After 7 days, influenza was detected by real-time polymerase chain reaction, immunohistochemistry, and viral culture in aortic plaques. The virus was sporadically detected in the blood, but not in other organs. Animals inoculated with respiratory syncytial virus as a control did not show dissemination of virus. The virus was associated with up-regulation of inflammatory cytokines and up-regulation of inflammatory gene expression. These findings demonstrate that viruses generally thought to be confined to the lung can translocate to areas of cardiovascular pathology.

The Role of Metabolomics Products

Metabolites reflect small molecules that are generated by cellular metabolic activities and represent the host genome, transcriptome, and proteome. Metabolomics measures byproducts of microbes and microbial communities most commonly in the GI tract, but potentially in other body sites as well. The effects of metabolomics on lung and heart disease may also arise from

metabolic products of inflammation stimulated by microbial communities. This area is just beginning to be explored for its relationship to infection and disease.

Links of the Metabolome to Cardiovascular Disease and GI Microbiota

Most work in this field has focused on the role of metabolic products of the GI microbiota on CVD. Red meat and egg consumption have been linked to CVD, and it has been hypothesized that commensal intestinal microbes could modify the interaction of diet with the host to alter this risk. A recent study examined the relationship of intake of phosphatidylcholine to trimethylamine-N-oxide (TMAO) in human volunteers fed hard-boiled eggs (a source of phosphatidylcholine) followed by blood and urine TMAO measurements (41). They were then treated with antibiotics, and blood and urine TMAO measurements were repeated. TMAO levels increased after egg consumption, but diminished after treatment with antibiotics, with a return to previous levels after antibiotic cessation. In a large cohort in the same study, TMAO levels were independent predictors of cardiovascular events (41). The relationship with metabolomic products, the microbiota, and infection is likely quite complex; a recent study in rats demonstrated that treatment with oral vancomycin increased infarct size after coronary artery ligation and that this effect could be diminished by

probiotics (42). These studies support the concept that the GI microbiota can interact with host factors to influence non-GI diseases and suggest novel therapeutic approaches.

Links of the Metabolome to Lung Disease

Little is known about the relationship of the metabolome to lung disease. There could be effects of changes in the lung metabolome or systemic metabolic products that could influence disease, but few studies have examined this relationship. A study of *Pseudomonas* strains from patients with cystic fibrosis showed different metabolites, and other studies of patients with cystic fibrosis have shown a relationship of sputum metabolites to lung function (43, 44), but much remains to be learned about metabolomics in lung disease.

The Role of Smoking

Smoking may influence the development of lung disease and CVD through modulation of infection. Studies have found alterations in the oral microbiome in smokers compared with nonsmokers, with increases in some species and decreases in others (45, 46). A recent study from the Lung HIV Microbiome Program found that oral microbial composition differed significantly by smoking status, but BAL communities in smokers and nonsmokers

were not significantly different (47). Smoking may also affect the GI microbiota. A study of smoking cessation reported that microbial populations found in stool samples clustered by smoking status and that samples taken after smoking cessation were distinguishable from those taken before smoking cessation and from current smokers (48).

Conclusions

Epidemiologic data support the coexistence of lung and cardiovascular diseases such as CVD and pulmonary hypertension. Infections are an attractive unifying cause or modifier of these diseases given the potential for direct or systemic effects of infectious organisms, their resulting inflammation, and/or metabolic compounds. These relationships are likely quite complex, with multiple routes between infection and disease possible. A better understanding of the links of infection to lung and heart disease will improve our understanding of the pathogenesis of these disorders and uncover novel therapeutic approaches. ■

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References

- Barnes PJ, Celli BR. Systemic manifestations and comorbidities of COPD. *Eur Respir J* 2009;33:1165–1185.
- Weitzenblum E, Chaouat A, Canuet M, Kessler R. Pulmonary hypertension in chronic obstructive pulmonary disease and interstitial lung diseases. *Semin Respir Crit Care Med* 2009;30:458–470.
- Kessler R, Faller M, Fourgaut G, Mennecier B, Weitzenblum E. Predictive factors of hospitalization for acute exacerbation in a series of 64 patients with chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 1999;159:158–164.
- Oswald-Mammoser M, Weitzenblum E, Quoix E, Moser G, Chaouat A, Charpentier C, Kessler R. Prognostic factors in COPD patients receiving long-term oxygen therapy: importance of pulmonary artery pressure. *Chest* 1995;107:1193–1198.
- van Gestel YR, Flu WJ, van Kuijk JP, Hoeks SE, Bax JJ, Sin DD, Poldermans D. Association of COPD with carotid wall intima-media thickness in vascular surgery patients. *Respir Med* 2010;104:712–716.
- Iwamoto H, Yokoyama A, Kitahara Y, Ishikawa N, Haruta Y, Yamane K, Hattori N, Hara H, Kohno N. Airflow limitation in smokers is associated with subclinical atherosclerosis. *Am J Respir Crit Care Med* 2009;179:35–40.
- McAllister DA, Maclay JD, Mills NL, Mair G, Miller J, Anderson D, Newby DE, Murchison JT, Macnee W. Arterial stiffness is independently associated with emphysema severity in patients with chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2007;176:1208–1214.
- Barr RG, Mesia-Vela S, Austin JH, Basner RC, Keller BM, Reeves AP, Shimbo D, Stevenson L. Impaired flow-mediated dilation is associated with low pulmonary function and emphysema in ex-smokers: the Emphysema and Cancer Action Project (EMCAP) Study. *Am J Respir Crit Care Med* 2007;176:1200–1207.
- Maclay JD, McAllister DA, Macnee W. Cardiovascular risk in chronic obstructive pulmonary disease. *Respirology* 2007;12:634–641.
- Sin DD, Wu L, Man SF. The relationship between reduced lung function and cardiovascular mortality: a population-based study and a systematic review of the literature. *Chest* 2005;127:1952–1959.
- Moro L, Pedone C, Scarlata S, Malafarina V, Fimognari F, Antonelli-Incalzi R. Endothelial dysfunction in chronic obstructive pulmonary disease. *Angiology* 2008;59:357–364.
- Chaouat A, Naeije R, Weitzenblum E. Pulmonary hypertension in COPD. *Eur Respir J* 2008;32:1371–1385.
- Fayngers V, Drakopanagiotakis F, Dennis McCool F, Klinger JR. Pulmonary hypertension in a stable community-based COPD population. *Lung* 2011;189:377–382.

- 14 Gan WQ, Man SF, Senthilselvan A, Sin DD. Association between chronic obstructive pulmonary disease and systemic inflammation: a systematic review and a meta-analysis. *Thorax* 2004;59:574–580.
- 15 Keatings VM, Barnes PJ. Granulocyte activation markers in induced sputum: comparison between chronic obstructive pulmonary disease, asthma, and normal subjects. *Am J Respir Crit Care Med* 1997;155:449–453.
- 16 Speich R, Jenni R, Opravil M, Pfab M, Russi EW. Primary pulmonary hypertension in HIV infection. *Chest* 1991;100:1268–1271.
- 17 Morris A, Gingo MR, George MP, Lucht L, Kessinger C, Singh V, Hillenbrand M, Busch M, McMahon D, Norris KA, et al. Cardiopulmonary function in individuals with HIV infection in the antiretroviral therapy era. *AIDS* 2012;26:731–740.
- 18 Drummond MB, Merlo CA, Astemborski J, Marshall MM, Kisalu A, McDyer JF, Mehta SH, Brown RH, Wise RA, Kirk GD. The effect of HIV infection on longitudinal lung function decline among injection drug users: a prospective cohort. *AIDS* 2013;27:1303–13011.
- 19 Drummond MB, Kirk GD, Astemborski J, Marshall MM, Mehta SH, McDyer JF, Brown RH, Wise RA, Merlo CA. Association between obstructive lung disease and markers of HIV infection in a high-risk cohort. *Thorax* 2012;67:309–314.
- 20 Crothers K, Huang L, Goulet JL, Goetz MB, Brown ST, Rodriguez-Barradas MC, Oursler KK, Rimland D, Gibert CL, Butt AA, et al. HIV infection and risk for incident pulmonary diseases in the combination antiretroviral therapy era. *Am J Respir Crit Care Med* 2011;183:388–395.
- 21 Almodovar S, Knight R, Allshouse AA, Roemer S, Lozupone C, McDonald D, Widmann J, Voelkel NF, Shelton RJ, Suarez EB, et al. Human immunodeficiency virus nef signature sequences are associated with pulmonary hypertension. *AIDS Res Hum Retroviruses* 2012;28:607–618.
- 22 Marecki JC, Cool CD, Parr JE, Beckey VE, Luciw PA, Tarantal AF, Carville A, Shannon RP, Cota-Gomez A, Tuder RM, et al. HIV-1 Nef is associated with complex pulmonary vascular lesions in SHIV-nef-infected macaques. *Am J Respir Crit Care Med* 2006;174:437–445.
- 23 Gingo MR, He J, Wittman C, Fuhrman C, Leader JK, Kessinger C, Lucht L, Slivka WA, Zhang Y, McMahon DK, et al. Contributors to diffusion impairment in HIV-infected persons. *Eur Respir J* (In press)
- 24 Sandler NG, Wand H, Roque A, Law M, Nason MC, Nixon DE, Pedersen C, Ruxrungtham K, Lewin SR, Emery S, et al.; INSIGHT SMART Study Group. Plasma levels of soluble CD14 independently predict mortality in HIV infection. *J Infect Dis* 2011;203:780–790.
- 25 Jiang W, Lederman MM, Hunt P, Sieg SF, Haley K, Rodriguez B, Landay A, Martin J, Sinclair E, Asher AI, et al. Plasma levels of bacterial DNA correlate with immune activation and the magnitude of immune restoration in persons with antiretroviral-treated HIV infection. *J Infect Dis* 2009;199:1177–1185.
- 26 Brenchley JM, Price DA, Schacker TW, Asher TE, Silvestri G, Rao S, Kazzaz Z, Bornstein E, Lambotte O, Altmann D, et al. Microbial translocation is a cause of systemic immune activation in chronic HIV infection. *Nat Med* 2006;12:1365–1371.
- 27 Yan AW, Schnabl B. Bacterial translocation and changes in the intestinal microbiome associated with alcoholic liver disease. *World J Hepatol* 2012;4:110–118.
- 28 Marchetti G, Bellistri GM, Borghi E, Tincati C, Ferramosca S, La Francesca M, Morace G, Gori A, Monforte AD. Microbial translocation is associated with sustained failure in CD4+ T-cell reconstitution in HIV-infected patients on long-term highly active antiretroviral therapy. *AIDS* 2008;22:2035–2038.
- 29 Douek D. HIV disease progression: immune activation, microbes, and a leaky gut. *Top HIV Med* 2007;15:114–117.
- 30 Ancuta P, Kamat A, Kunstman KJ, Kim EY, Autissier P, Wurcel A, Zaman T, Stone D, Mefford M, Morgello S, et al. Microbial translocation is associated with increased monocyte activation and dementia in AIDS patients. *PLoS ONE* 2008;3:e2516.
- 31 Balagopal A, Philp FH, Astemborski J, Block TM, Mehta A, Long R, Kirk GD, Mehta SH, Cox AL, Thomas DL, et al. Human immunodeficiency virus-related microbial translocation and progression of hepatitis C. *Gastroenterology* 2008;135:226–233.
- 32 Lozupone C, Cota-Gomez A, Palmer BE, Linderman DJ, Charlson ES, Sodergren E, Mitrev A, Abubucker S, Martin J, Yao G, et al.; Lung HIV Microbiome Project. Widespread colonization of the lung by *Tropheryma whippelii* in HIV infection. *Am J Respir Crit Care Med* 2013;187:1110–1117.
- 33 Copeland DR, McVay MR, Dassinger MS, Jackson RJ, Smith SD. Probiotic fortified diet reduces bacterial colonization and translocation in a long-term neonatal rabbit model. *J Pediatr Surg* 2009;44:1061–1064.
- 34 Huang L, Crothers K, Morris A, Groner G, Fox M, Turner JR, Merrifield C, Eiser S, Zucchi P, Beard CB. *Pneumocystis* colonization in HIV-infected patients. *J Eukaryot Microbiol* 2003;50:616–617.
- 35 Morris A, Kingsley LA, Groner G, Lebedeva IP, Beard CB, Norris KA. Prevalence and clinical predictors of *Pneumocystis* colonization among HIV-infected men. *AIDS* 2004;18:793–798.
- 36 Morris A, Alexander T, Radhi S, Lucht L, Sciruba FC, Kolls JK, Srivastava R, Steele C, Norris KA. Airway obstruction is increased in *Pneumocystis*-colonized human immunodeficiency virus-infected outpatients. *J Clin Microbiol* 2009;47:3773–3776.
- 37 Desmet S, Van Wijngaerden E, Maertens J, Verhaegen J, Verbeken E, De Munter P, Meersseman W, Van Meensel B, Van Eldere J, Lagrou K. Serum (1-3)-beta-D-glucan as a tool for diagnosis of *Pneumocystis jirovecii* pneumonia in patients with human immunodeficiency virus infection or hematological malignancy. *J Clin Microbiol* 2009;47:3871–3874.
- 38 Watanabe T, Yasuoka A, Tanuma J, Yazaki H, Honda H, Tsukada K, Honda M, Gatanaga H, Teruya K, Kikuchi Y, et al. Serum (1→3)-beta-D-glucan as a noninvasive adjunct marker for the diagnosis of *Pneumocystis* pneumonia in patients with AIDS. *Clin Infect Dis* 2009;49:1128–1131.
- 39 Morris A, Hillenbrand M, Finkelman M, George MP, Singh V, Kessinger C, Lucht L, Busch M, McMahon D, Weinman R, et al. Serum (1→3)-beta-D-glucan levels in HIV-infected individuals are associated with immunosuppression, inflammation, and cardiopulmonary function. *J Acquir Immune Defic Syndr* 2012;61:462–468.
- 40 Haidari M, Wyde PR, Litovsky S, Vela D, Ali M, Casscells SW, Madjid M. Influenza virus directly infects, inflames, and resides in the arteries of atherosclerotic and normal mice. *Atherosclerosis* 2010;208:90–96.
- 41 Tang WH, Wang Z, Levison BS, Koeth RA, Britt EB, Fu X, Wu Y, Hazen SL. Intestinal microbial metabolism of phosphatidylcholine and cardiovascular risk. *N Engl J Med* 2013;368:1575–1584.
- 42 Lam V, Su J, Koprowski S, Hsu A, Tweddell JS, Rafiee P, Gross GJ, Salzman NH, Baker JE. Intestinal microbiota determine severity of myocardial infarction in rats. *FASEB J* 2012;26:1727–1735.
- 43 Behrends V, Ryall B, Zlosnik JE, Speert DP, Bundy JG, Williams HD. Metabolic adaptations of *Pseudomonas aeruginosa* during cystic fibrosis chronic lung infections. *Environ Microbiol* 2013;15:398–408.
- 44 Yang J, Eiserich JP, Cross CE, Morrissey BM, Hammock BD. Metabolomic profiling of regulatory lipid mediators in sputum from adult cystic fibrosis patients. *Free Radic Biol Med* 2012;53:160–171.
- 45 Charlson ES, Chen J, Custers-Allen R, Bittinger K, Li H, Sinha R, Hwang J, Bushman FD, Collman RG. Disordered microbial communities in the upper respiratory tract of cigarette smokers. *PLoS ONE* 2010;5:e15216.
- 46 Shchipkova AY, Nagaraja HN, Kumar PS. Subgingival microbial profiles of smokers with periodontitis. *J Dent Res* 2010;89:1247–1253.
- 47 Morris AB, Beck JM, Schloss PD, Campbell TB, Crothers K, Curtis JL, Flores SC, Fontenot AP, Ghedin E, Huang L, et al.; Lung HIV Microbiome Project. Comparison of the respiratory microbiome in healthy nonsmokers and smokers. *Am J Respir Crit Care Med* 2013;187:1067–1075.
- 48 Biedermann L, Zeitz J, Mwinyi J, Sutter-Minder E, Rehman A, Ott SJ, Steurer-Stey C, Frei A, Frei P, Scharl M, et al. Smoking cessation induces profound changes in the composition of the intestinal microbiota in humans. *PLoS ONE* 2013;8:e59260.