The task of a meeting summarizer is to provide a perspective for the proceedings of the meeting, and to some extent, portend where the field will be when the next pulmonary circulation conference is held several years from now. The first task is much easier than the latter, but several new, emerging directions were evident at this conference, making it possible to predict the issues and concepts that will be addressed in the near future. Thanks go to Norbert Voelkel and the conference committee for organizing what truly was a state-of-the-art look into the "Biology & Pathology of the Lung Circulation." Thanks also to the speakers and conference participants who made the meeting a provocative discussion of the important questions facing the field.

For many years, the study of the pulmonary circulation has been considered a less mature cousin of its systemic relative. With this meeting, however, it is evident that the lesser cousin has grown up and has much to teach us. The factors that have made the study of the pulmonary circulation so difficult—its cellular and functional diversity—are now being used to the investigator's advantage to understand mechanisms of vascular development, physiology, and remodeling. In many instances, experimental systems have been developed and exploited in pulmonary vessels that are difficult or impossible to model in the systemic circulation. Yet, many of the basic principles that have been learned from these efforts apply to both vascular beds.

In looking back 10 years to the last Aspen Lung Conference on pulmonary hypertension,¹ the concept of cellular heterogeneity had been introduced with the finding of multiple populations of smooth muscle cells (SMC) present in the media of normal and hypertensive vessels. This newly described complexity challenged our conventional thinking that all cells within a vascular compartment were the same. There had been hints from work in the systemic circulation that "subpopulations" of SMCs could selectively respond to various injurious stimuli, but the extent of the cellular heterogeneity only became evident when the synthetic phenotype was evaluated with probes for production of various matrix proteins. Then, there was much discussion and speculation about what this heterogeneity meant to processes such as vascular development and remodeling, and now, 10 years later, we are beginning to gain some answers to those questions.

Few would argue with the suggestion that the endothelial cell may be the most important "playmaker" in the early vascular response to injury. Through its ability to sense changes in pressure, flow, and oxygen tension, and to produce potent messenger molecules that provide instructive signals to other vascular cells, the endothelial cell may be the most important player in initiating the remodeling events that lead to alterations in vascular tone and flow patterns in the lung. But the endothelial cell is more than just a sensor/signaling unit in that it can also undergo dramatic changes in proliferative state, leading to obstructive pathologic condition. Dr. Rubin Tuder has shown that cells within the neointima of the plexiform lesion display markers characteristic of endothelial cells. Since plexiform lesions are found most often at the periphery of the lung, these important results raise interesting questions about why there are segmental differences in the endothelial cell response. Is there a phenotypic heterogeneity in endothelial cells like that demonstrated for SMCs, or is the response dictated partially or in its entirety by physiological factors? A partial answer to this question was provided also by Dr. Tuder's laboratory showing that endothelial cells in the lesions of patients with primary pulmonary hypertension (PPH) were monoclonal, that is, derived from a single or very limited number of progenitor or "stem" cells. In contrast,
endothelial cells in lesions from patients with secondary pulmonary hypertension were polyclonal, essentially derived from a general expansion of the endothelial cell population at that site. The difference between the two pathologic conditions is an important one: monoclonal expansion implies neoplasia-like growth, whereas polyclonal expansion implies the presence of chemical mediators, such as those associated with inflammation, that alter cell proliferation generally. A more detailed definition of the two mechanisms, and the cellular events responsible for their activation, promises to be one of the more exciting lines of research in endothelial cell biology.

Monoclonality associated with PPH is worthy of special attention because it provides us with a mechanistic explanation for the origins and expansion of cells in chronic lesions. Another selection mechanism of equal importance, however, is the activation or suppression of specific subpopulations of cells based on their ability to express a specific phenotype. In this case, gene regulation and not cell proliferation is the operative mode.

Using a panel of antibodies to SMC marker proteins, Dr. Maria Frid and colleagues, and Dr. Kurt Stenmark identified several unique cell populations in the vascular media of both adult and neonatal bovine pulmonary arteries. These cell populations show differences in morphology, growth, cytoskeletal/contractile proteins, and extracellular matrix synthesis. The stability of the cell phenotypes was shown by Dr. Frid and coworkers who were able to isolate two populations of cells from the vascular media. One, termed “immature,” expressed little smooth muscle-actin and no smooth muscle-myosin but exhibited rapid growth. A second population, called “differentiated,” contained slowly dividing cells that expressed both smooth muscle marker proteins. When exposed to hypoxia, the proliferative potential of the immature cells but not the differentiated cells was enhanced. Interestingly, hypoxia stimulated the secretion of a mitogenic factor by the immature cells that could augment the proliferative response of the differentiated cells. The importance of this observation to vascular repair is obvious and the complexity somewhat disheartening. We must conclude from these findings that subpopulations of cells exist in the vessel wall that, in response to specific stimuli such as hypoxia, are capable of proliferating in the absence of mitogens and of secreting factors that can act in a paracrine fashion to induce proliferation or alter gene expression of neighboring cells. They also suggest that not all cells in the vascular wall respond to a hypertensive stimulus in the same way.

Dr. Edward Dempsey and colleagues outlined how some SMCs require a protein kinase C (PKC)-dependent priming step in order to proliferate in response to hypoxia. Similarly, Dr. Mark Gillespie and coworkers found that differences in polyamine transport can discriminate between two populations of cultured rat SMCs that differ in terms of their morphology and proliferative rate in response to hypoxia.

I agree with Dr. Stenmark’s suggestion that this cellular diversity exists to address the various biological functions required of SMCs in both normal and pathologic conditions. For example, in response to pathologic stimuli, some medial SMCs are likely to maintain their normal, mainly contractile functions, whereas others are likely to exhibit elevated proliferation and/or matrix protein synthesis. This diversity in cellular responses would lend itself to better maintenance of vascular homeostasis after vascular injury. Most functions could continue without the severe compromise that would occur if all (or most) cells shifted simultaneously from one phenotype (eg, contractile) to another (proliferative and/or synthetic). Importantly, the heterogeneity in response to various “trigger factors” suggests that some SMCs might have signaling function while others respond to the signals they generate.

As might be expected, the effects of hypoxia were extensively discussed during the meeting which brings up questions that were raised 10 years ago for which we now have a more mechanistic answer. What are the oxygen sensors on the cell, and how does the cell sense hypoxia? Dr. E. Kenneth Weir reported on his work showing that entry of calcium into the cell is controlled by ion channels that act to control the distribution of charged ions across the cell membrane. Under resting, normoxic conditions, there is an outward flow of positively charged potassium ions through voltage-dependent potassium channels, that work to oppose membrane depolarization. If these channels are blocked, vasoconstriction results as a consequence of entry of extracellular calcium into the cytosol. Interestingly, an increase in cytoplasmic calcium activates a second type of potassium channel, called large-conductance channels, that facilitates an outward flow of potassium and repolarization of the membrane.

Hypoxic vasoconstriction involves inhibition of potassium current, membrane depolarization, and calcium entry through calcium channels. The oxygen-sensing mechanism is thought to depend on the membrane sulfhydryl redox status, related in turn to reduced generation of oxygen radicals during hypoxia. The specific cellular response to oxygen, however, once again illustrates the concept of phenotypic heterogeneity mentioned above: Hypoxic vasoconstriction is unique to pulmonary SMCs and occurs predominantly in small pulmonary arteries (200 to 300 μm). Depending on the developmental age, vascular segment, and animal species, the oxygen-sensitive potassium channel can be either the voltage-dependent potassium or large-conductance channels. Even the L-type calcium channels have been shown to be oxygen-sensitive in some cells.

In addition to hypoxia, vasoconstriction could result from the increased production of vasoconstrictors such thromboxane A2, endothelin, and 5-hydroxytryptamine, or from the decreased production of endogenous dilators such as nitric oxide or prostacyclin. Ultimately, all of these agents alter vascular tone by altering concentrations of cytoplasmic calcium either through release of intracellular stores or by entry of extracellular calcium, primarily through the L-type calcium channels. Other channels are also important in maintaining intracellular potassium levels. Dr. Sharon Rouns and colleagues showed that oxidant injury of endothelial cells stimulated the sodium-potassium ATP pump which is important in maintaining intracellular/extracellular Na and K gradients by pumping Na out and K into the cell. Changes in Na/K pump activity
are likely important in the maintenance of endothelial cell homeostasis after oxidant stress.

The study of hypoxic pulmonary vasoconstriction has taught us much about how ion flux mediates vascular tone, but the story is more than just cells stretching and contracting. As discussed above, many cells alter their proliferative or synthetic properties in response to hypertensive injury, and recent studies have shown that many of these changes are in direct response to hypoxia. As a result, there has been great interest in the identification of genes responsible for the hypoxic response. There may be several candidates for this role depending on the differentiation state of the vessel and the cell type, but one factor that has recently moved into the spotlight is HIF-1.

HIF-1 was originally identified as a nuclear factor that was induced by hypoxia and bound to a site in the erythropoietin gene hypoxia-response element. Dr. Gregg Semenza reported that HIF-1 now appears to be an important transcription factor for several genes whose products mediate essential cellular responses to hypoxia, including erythropoiesis, glycolysis, and vasodilatation. Interestingly, Dr. Maria Frid and coworkers found that HIF was upregulated in the vascular wall immature cell population in response to hypoxia. Similarly, upregulation of inducible nitric oxide synthase (iNOS) in response to hypoxia occurs as a consequence of HIF-1 interacting with a promoter element in the synthase gene (Dr. Lisa Palmer and colleagues). Regulation of eNOS and preproET-1 expression in response to hypoxia (Dr. Timothy Le Cras and coworkers) may also occur via a HIF-1 regulated pathway.

Like other transcription factors that serve a protective function in the cell, the activity of HIF-1 is carefully regulated. The active HIF-1 transcription factor is actually made up from two distinct gene products, HIF-1α and HIF-1β. Both proteins are rapidly translocated into the nucleus in response to hypoxia where they associate to from a heterodimer which is the active product. Upon return to normoxia, nuclear HIF-1 is rapidly degraded. (Note: the HIF-1α gene was mapped to human chromosome 14, which excludes it as a direct candidate for the familiar hypertension gene located on chromosome 2, see below). It will be of great interest to learn which genes are regulated by HIF-1 and the cellular consequences of their regulation. Also of great interest will be the delineation of how the HIF-1α and HIF-1β genes themselves are regulated by oxygen tension.

It is now clear that PPH in the adult and pulmonary hypertension in the neonate are two distinct disease processes with differing pathologies, and in many instances, differing clinical outcomes. If the primary cause of pulmonary hypertension is treated quickly, there is a good chance that the disease will regress. This is most often the case in children where heart defects, for example, can be detected early and surgically repaired. If the primary cause is unknown or detected late in the progress of the disease, such as in adults with PPH, the disease is usually fatal. Progress has been made, however, in the treatment of unexplained PPH through lung transplantation and through the continuous infusion of prostacyclin. Interestingly, prostacyclin infusion has dramatically reduced symptoms and pulmonary artery resistance despite lack of acute hemodynamic effects. More should be learned about the effects of prostacyclin from animal studies involving overexpression of prostacyclin synthase. In an isolated perfused lung model, Dr. Mark Geraci and colleagues have shown that acute transfection of prostacyclin synthase resulted in reduced hypoxic vasoconstriction compared with lungs transfected with a reporter construct. This group has also generated transgenic mice with prostacyclin overproduction. The phenotype of these mice, and how they respond to hypertensive stimuli will undoubtedly help clarify the chronic effects of PGI2 production on structural remodeling of the pulmonary circulation.

An exciting recent advance that has focused the attention of the field in great anticipation is the linkage of a familial form of PPH to a locus within a 27 cM region of chromosome 2 (between 2q31-2q32) (Dr. Jane Morse and colleagues). The large region of involvement and a phenotype typical of incomplete penetrance suggests that multiple genes within this region may be involved. By the time of the next Aspen Conference on pulmonary hypertension, the genetic defect at the PPH1 locus will most likely be elucidated and provide the first look into genes, and hence, mechanisms of pulmonary vascular remodeling.

In nongenetic forms of adult PPH, results from several studies presented at the meeting strongly suggest that vascular remodeling is a cellular response to injury, perhaps most similar to cellular activation in wound repair or, as suggested by Dr. Tudor’s findings, a process involving neoplastic-like expansion. The lesion in neonatal pulmonary hypertension, in contrast, is an alteration of the normal developmental program through which the pulmonary vascular cells are progressing. A major impediment to understanding the cellular mechanisms of vascular remodeling in adult PPH has been the absence of an animal model that replicates all of the pathologic findings in the human disease. Whereas neointimal formation is prominent in PPH, animal models of vessel injury, such as monocrotaline (MCT), demonstrate predominantly medial changes. It has been known for some time that either injury or altered hemodynamics is associated with vascular remodeling in systemic arteries, but it was not clear how these two factors relate to each other in either the initiation or progression of the disease state in PPH. An answer to this question was provided by Dr. Mitchell Botney and coworkers who developed a rat model of vascular injury in which MCT followed left pneumonectomy. Within a relatively short period, these animals developed neointimal lesions that resembled those found in the human disease. Neointimal lesions did not form in response to injury alone (MCT) or flow without injury (pneumonectomy without MCT).

These findings, as well as information from other disease models, suggest that the vascular changes associated with PPH result from a multistep process driven by both injury and hemodynamic changes. One scenario proposed by Dr. Botney is that the initial response to “injury” is medial hypertrophy leading to altered hemodynamics, which in turn, results in neointimal changes. If this
is true, then what is the “injury” that occurs in humans to
initiate the hemodynamic changes associated with PPH? A
possible link between viral infection or exposure to toxins
and vascular changes leading to PPH is intriguing but
requires more work to show true cause and effect. Access
to human PPH tissue from lung transplant programs now
gives us the opportunity to investigate such questions.

Analysis of the possible injurious mechanisms must also
include thrombosis as a early event in vascular cell activa-
tion. The presence of thrombin activity in the vessel wall is
of special interest because many important pathways and
effect molecules may be activated or deactivated via
thrombin’s proteolytic properties. Moreover, there is evi-
dence that nonproteolytic domains of thrombin have
biological activity and can directly modulate gene tran-
scription of vasoactive substances. Dr. Gary Visner and
colleagues showed us that the gene for endothelin-1 is
stimulated in endothelial cells by thrombin through mod-
ulation of trans-acting factors acting on a specific region of
the promoter. Thrombin also can act directly on smooth
muscle cells as a vasoconstrictor. These properties suggest
an important dual function of thrombin in inducing con-
striction of small vessels at sites of injury while neighbor-
ing, undamaged vessels are stimulated to increase their
blood flow. Tissue factor is another agent that seems to
play an important role in vascular remodeling. A trans-
membrane molecule that can initiate clotting in vitro,
tissue factor was shown by Dr. David Stern and colleagues
to be expressed by monocytes in hypoxic mice. Interest-
ingly, PAI-1 was also upregulated in mononuclear cells in
response to hypoxia, whereas tissue plasminogen activator
and urokinase were downregulated. It is also interesting
that the monocyte chemoattractant MCP-1 is upregulated
in response to hypoxia and that MCP-1 -/- mice show no
fibrin deposition. Together, these findings suggest that the
monocyte may be a more important player in thrombotic/
coagulative events in vascular remodeling than previously
thought.

Thrombosis is not the whole story with inflammation,
however. Dr. Stern and colleagues have provided interesting
insight into the role of inflammation in tissue remodel-
ing based on their studies of preservation of lung transplant
tissue. They showed that a key molecule in-
duced during hypoxia is P-selectin, an adhesion molecule
on the surface of endothelial cells that recognizes specific
oligosaccharide chains on the surface of neutrophils. In
lung preservation studies, tissue from mice that were
deficient in P-selectin showed enhanced survival due to
decreased leukocytes in the tissue. This finding raises an
important issue that was only briefly addressed at the
meeting but will receive more attention in years to come:
namely, the role of adhesion molecules in the targeting
and extravasation of inflammatory cells or in the move-
ment of vessel wall cells from one compartment to
another. The data of Dr. Stern and colleagues indirectly
suggest that agents capable of blocking cellular migration
or adhesion may help ameliorate further lesion formation
at sites of vascular injury. Learning more about adhesion
receptors will also be important in understanding how
cells adhere to their extracellular matrix substrate and
sense frictional forces that result in transduction of bio-
chemical messages responsible for regulating matrix pro-
duction (Dr. Jill Bishop and colleagues) and vascular tone.
Dr. Abdul Barakat and colleagues presented fascinating
work showing that shear-induced responses depend on the
precise shear stress distribution on the endothelial cell
surface. Signals in response to changes in flow are gener-
ated by flow-mediated ion channels and reorganization of
the cytoskeleton. It is safe to assume that adhesion
receptors that anchor the cell to the basement membrane
will turn out to be major signaling agents in flow sensing.
This undoubtedly is an area that will attract a lot of
attention in future years as adhesion receptors and their
ligands become better characterized.

Pattern formation in biology has fascinated scientists
with interests as diverse as mathematicians and develop-
mental biologists. The application of mathematical mod-
eling to the fractal branching pattern of the pulmonary
circulation is an example where the development of new
scientific tools allows us to reinvestigate old questions. In
this case, the question that was revisited concerned the
blood flow distribution in the lung. By applying rules of
fractal geometry to blood flow patterns, Dr. Robb Glenny
found that pulmonary blood flow was neither uniform nor
random. High flow segments stayed high flow, low flow
stayed low flow, implying that flow is determined by
arterial geometry and does not change over time. Even
when flow was increased by exercise, the perfusion pattern
remained the same. The heterogeneity observed for per-
fusion was also evident for ventilation, with a matching of
high flow and high perfusion regions. Taking a similar
fractal approach to study perfusion in subpleural alveolar
walls, Dr. Wiltz Wagner and colleagues found that the
pattern of flow switching from one capillary segment to
another followed a predicted fractal relationship. Fur-
thermore, there was no relationship between perfusion of the
capillary network in adjoining alveolar walls, which sug-
gests that if one alveolar wall is disrupted, the neighbors
would not be affected. In addition to teaching us more
about the perfusion properties of the lung, these studies
highlight an important question about lung development:
What drives the fractal development of the pulmonary
circulation? To answer this question, we need to know
more about the developmental signals that control the
differentiation and spatial positioning of cells. How do
cells know where to form bifurcations or when to activate
specific genes associated with their differentiated pheno-
type? Several studies have established that the developing
airways play a major role in providing directional and,
most likely, differentiation signals for the developing
vasculature. The studies of Dr. Glenny and Dr. Wagner
and colleagues present a compelling argument for why
that needs to be.

Many people are willing to try any miracle diet or drug
that will take off a few pounds. The recent increase in
pulmonary hypertension associated with the use of the
appetite suppressant drugs fenfluramine, fenfluramine
(the combination of the two drugs popularly known as
“fen-phen”), and dexfenfluramine, however, once again
tells us that there is no such thing as a painless approach
to weight loss. The evidence for an increased risk of
pulmonary hypertension in people (particularly young

CHEST / 114 / 1 / JULY, 1996 SUPPLEMENT 109S
women) taking these anorexic drugs is incontrovertible. In an international study of pulmonary hypertension (IPPHS) coordinated by Dr. Lucien Abenhaim, the mortality and survival of patients taking fenfluramine was as bad as patients with classic PPH. Even more worrisome was the finding that discontinuing the drug did not stop the progression of the disease. Between the time of the Aspen Lung Conference in June 1997 and the writing of this summary, the manufacturer of fenfluramine and dexfenfluramine, at the FDA’s request, withdrew the drugs from general use because of an association with valvular heart disease. While this is clearly a welcome and long overdue step, the continued progression of the disease after drug cessation means that physicians will continue to see anorexigen-associated cases of PPH for years to come. All pulmonary and critical care physicians should become familiar with the data surrounding the fenfluramines and continue to be cognizant of possible associations between the intake of appetite-depressant drugs and hypertensive disease.

As a consequence of our attempts to understand how the fenfluramines work, we have gained an intriguing glimpse into another cellular mechanism associated with hypertensive disease. Dexfenfluramine acts through the following four mechanisms: (1) it increases serotonin-mediated neurotransmission by blocking the uptake of serotonin; (2) its principal metabolite, dexnorfenfluramine, directly releases serotonin into synapses and (3) activates the serotonin 5HT2 receptors; and (4) dexfenfluramine also blocks K channels, leading to increased cytosolic calcium. Somewhere hidden among these diverse effects is an important clue about the etiology of PPH. It is interesting that the valvular problems which led to the withdrawal of fenfluramine from the market were often of a specific type associated with serotonin-excess conditions.

I have taken little space so far describing chemical mediators of vasoconstriction, which may be a surprise for a meeting on pulmonary hypertension. While this undoubtedly reflects my bias towards cell biology, it also accurately reflects the emphasis of the meeting and the research direction of the field. Initially considered a disease of vasoconstriction, it is clear from the presentations at this conference that medial and adventitial hypertrophy and neointimal formation are the key elements explaining the pathophysiology of the disease. As our understanding of the mechanisms of remodeling improve, it is becoming apparent that many of the same factors that regulate vascular tone also participate in remodeling. Endothelin-1, a potent vasoconstrictor, and angiotensin II, for example, are increased in pulmonary arteries of hypertensive human subjects and are known to stimulate cell proliferation and expression of extracellular matrix proteins. ACE expression is also increased at sites of active remodeling as is transforming growth factor-beta (TGF-β). TGF-β stimulates production of several vascular matrix proteins and stimulates SMC replication. Although TGF-β has no direct effect on vasoconstriction, it does inhibit nitric oxide synthase and stimulates endothelin and angiotensin II production.

In contrast to the vasoconstrictors, nitric oxide synthase is decreased in hypertensive vessels. Although a direct role of NO in vascular remodeling has not been demonstrated, it does inhibit expression of several of the mediators described above, including endothelin-1 and angiotensin II. It is interesting that mice deficient in endothelial NO synthase (NOS3) develop mild pulmonary hypertension consistent with a role for NO in maintaining the normal low pulmonary vascular tone in the mouse lung (Dr. Timothy Quinlan and colleagues). However, the main pulmonary artery of the NOS3-deficient mice, which was morphologically similar to the NOS3-sufficient animal, did not undergo vasoconstriction in response to acetylcholine, suggesting that NO may play some role in vascular cell differentiation. Another vasodilator that is rapidly produced by SMCs during acute hypoxia is carbon monoxide (CO) (Dr. Stella Kourembanas). Like the other factors mentioned above, CO can have a direct effect on remodeling by suppressing vascular endothelial growth factor (VEGF), endothelin-1, and platelet-derived growth factor-B (PDGF-B). The effects of CO are transient, however, occurring over a period of approximately 48 h. This suggests a completely new mechanism for acute regulation of lung injury.

The renin-angiotensin system is worthy of special attention because of its diverse effects on vascular tone, hormone secretion, and tissue growth. Work reported by Dr. Nicholas Morrell and coworkers showed that angiotensin II stimulates the proliferation of SMCs via the AT1 receptor, which is the major receptor for mediating many of angiotensin’s biological effects. Recently, however, a second receptor subtype called AT2 has been characterized which operates through a different signaling pathway. Work by Dr. Victor Dzau’s group showed that the AT2 receptor is widely expressed in fetal tissues but present only at limited levels in adult tissues. The AT2 receptor has antiproliferative properties and is capable of offsetting the growth promoting effects mediated by the AT1 receptor through a very interesting pathway: AT2 receptor stimulation activates mitogen-activated protein (MAP) kinase phosphatase-1 which inhibits MAP kinase activation and phosphorylation of Bcl-2, resulting in the induction of apoptosis. These results support the possibility that AT2 receptor plays an important role in fetal development and in the pathogenesis of some diseases in which apoptosis is involved. This has been shown most convincingly by in vitro studies where transfer of the rat AT2 receptor into an injured rat carotid artery resulted in attenuation of neointimal hyperplasia. Not only are these studies important in understanding the role of angiotensin and its receptors, but they also illustrate the potential that in vivo gene transfer technology provides for disease therapy.

In his summary of the last conference 10 years ago, Dr. Jack Reeves predicted that an understanding of the pathophysiology of pulmonary hypertension would require an understanding of cellular and molecular mechanisms of vascular remodeling. The clear message from this present meeting is that is exactly where the field is concentrating. Jack also pointed out, however, that what was missing was a “unifying theory” to guide the field. Have we made
progress in understanding and treating pulmonary hypertension? The answer is a resounding yes, but as a participant in the last conference, I think that there are just as many unanswered questions today as 10 years ago, only the questions are different. Perhaps it is impossible to develop a unifying theory in a field that touches on as many different disciplines as does the pulmonary circulation. The nature of scientific inquiry, in fact, requires flexibility and change as new data become available. It is only when the ideas are the same after 10 years that one has to question scientific progress. Fortunately, we have moved beyond that. We still wait, however, for the “moment when the wind drops, the surface of the pool clears and is stilled, and one sees deeply with limpid simplicity into the nature of things” (Horace Freeland Judson, in The Eighth Day of Creation).

REFERENCE