Materials Science

Now You See Them

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Crystallization lies at the heart of many natural and technological processes, from the production of pharmaceuticals and nanomaterials to the formation of bones and teeth, frost heave, and scale deposition. Crucial features of these crystals, such as lattice orientation, particle size, and size distribution, are defined by conditions during the earliest stages of precipitation—at nucleation. Yet, nucleation from solution is poorly understood, because experimental studies of nucleation are highly challenging (1). Recent studies have highlighted the possible role of clusters in nucleation within precipitation (2, 3). On page 1819 of this issue, Gebauer et al. provide support for this thesis (4) by demonstrating the presence of large, well-defined clusters before nucleation of one of the phases of calcium carbonate. Crystallization appears to proceed through aggregation of these clusters. The results challenge the conventional picture of crystal nucleation.

Classical nucleation theory provides a simple understanding of how crystals nucleate. Nucleation is often slow because of a free-energy barrier originating from the interface between the nucleus and its surroundings. The theory assumes that nuclei grow one molecule at a time (see the figure, top). Gebauer et al. now suggest a different mechanism, in which nucleation of ACC occurs by aggregation of stable, amorphous, precritical clusters (bottom). The nucleated ACC phase subsequently crystallizes to generate the final stable crystal product.

**How do crystals nucleate?** According to classical nucleation theory, calcium carbonate nucleation proceeds by addition of ions to a single cluster (top). Gebauer et al. now suggest a different mechanism, in which nucleation of ACC occurs by aggregation of stable, amorphous, precritical clusters (bottom). The nucleated ACC phase subsequently crystallizes to generate the final stable crystal product.

Can new results on calcium carbonate nucleation be reconciled with classical nucleation theory?

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**CLASSICAL THEORY**

- Reversible addition of ions to precritical cluster
- Nucleation
- Postcritical nucleus
- Growth

**ALTERNATIVE MECHANISM**

- Formation of stable precritical clusters
- Aggregation
- Nucleation of crystalline phase
- Growth
- Final crystal

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saturate at which nucleation occurs is determined largely by the size of the nanodroplets present (10).

Could the precritical clusters observed by Gebauer et al. also be the result of stabilization of calcium carbonate clusters by another species present as an impurity? This mechanism would provide a basis for stabilizing precritical clusters in a free-energy minimum and does not contradict classical nucleation theory. Such impurities are ubiquitous and virtually impossible to eliminate from any solution. The results of Gebauer et al. may thus reflect the mechanism of nucleation of calcium carbonate in “real” systems. Nucleation could then occur by coalescence of the precritical clusters to give ACC, which will subsequently crystallize to a more stable crystalline polymorph. The latter mechanism is consistent with the observations of Gebauer et al., who show that ACC is the first phase precipitated after nucleation.

The idea that nucleation of calcium carbonate may proceed via an aggregation mechanism is highly topical. The past decade has seen great progress in understanding crystallization processes, and it is now well recognized that single-crystal growth (as distinct from nucleation) often occurs via the aggregation of small precursor units rather than by addition of ions or molecules to a nucleus (11). Cluster species have also been observed before nucleation in saturated solutions of compounds such as sodium chloride (2), urea (2), and glycine (3), and there have been suggestions that clustering can determine which polymorph is formed (13). However, none of these even remotely approach the size or stability of the clusters observed by Gebauer et al. Further investigation of precritical clusters and their role in the crystallization of calcium carbonate, and indeed other compounds, is eagerly anticipated.

References

Human genetic studies have led to the identification of a transcriptional regulator that could serve as a therapeutic target for adult hemoglobin disorders.

PREVIOUS PAGE

发育生物学

从基因关联到基因开关

Alan M. Michelson

 deciphering the sequence of the human genome and the subsequent cataloging of common human DNA sequence variation marked a paradigm shift in human genetics. These resources, together with advances in cost-effective genotyping technologies, enabled the design of genome-wide association studies for the unbiased discovery of commonly occurring DNA sequence variations called single-nucleotide polymorphisms (SNPs) that are preferentially associated with a disease or other clinical trait (1). Although genome-wide association studies have uncovered disease-associated SNPs, identifying actual disease-causing variants—and gaining deep insights into how those variants generate the underlying molecular pathophysiology—have so far yielded only modest results. This has led to criticisms of the genome-wide association approach for investigating the etiologies of common diseases (2). However, this assessment may be premature. On page 1839 of this issue, Sankaran et al. (3) show how genome-wide association findings can lead to a detailed understanding of disease mechanisms and be used to ascertain novel therapeutic targets.

Previous genome-wide association studies conducted in independent populations have identified SNPs in three chromosomal loci that are associated with varying expression levels of human fetal hemoglobin (HbF) (4–6). HbF is a clinically important quantitative trait because elevated concentrations reduce the severity of sickle cell disease and β-thalassemia, disorders caused by different mutations in the human β-globin gene (7). Normally, HbF predominates in the fetus but declines to very low amounts postnatally due to repression and activation of the γ-globin and β-globin genes, respectively. The “switch” that controls these reciprocal changes in globin gene expression has been extensively investigated, but the molecular basis of this developmental process remains largely unknown. As reported by Sankaran et al., functional studies motivated by recent HbF genome-wide association findings have provided a major breakthrough in understanding the hemoglobin switching problem.

One of the SNPs associated with elevated HbF expression is found in an intron (noncoding region) of the BCL11A gene on human chromosome 2 (see the figure). BCL11A encodes a protein that represses transcription in the B lymphoid lineage (8).

Sankaran et al. hypothesized that BCL11A might repress expression of the γ-globin gene, with expression or activity of this repressor correlating inversely with HbF production both during normal development and in individuals of different genotypes at the BCL11A locus. They first determined that BCL11A is expressed as two long isoforms, encoded by alternatively spliced messenger RNAs (mRNAs), in primary adult erythroblasts. By contrast, only shorter variants of BCL11A are found in human embryonic erythroleukemia cells and in primary human fetal liver cells, both of which express high amounts of HbF. Moreover, the genotype at the BCL11A SNP that affects HbF production influences expression of mRNAs encoding the long isoforms in lymphoblastoid cell lines: High expression of BCL11A mRNA corresponds to homozygosity for the allele associated with low HbF production; low mRNA expression corresponds to homozygosity for the allele associated with high HbF production; and SNP heterozygotes express intermediate amounts of mRNA (see the figure). If the association between the BCL11A SNP and the expression level of this gene in lymphobla-