functions as an inhibitor of the spindle assembly checkpoint, called p31^{comet} [18]. Interestingly, p31^{comet} only binds to C-Mad2 [19] and can compete with O-Mad2 for binding to C-Mad2 [17], indicating that it might inhibit the spindle assembly checkpoint by preventing formation of the Mad2 dimer. Yang *et al.* now described the crystal structure of p31^{comet} bound to C-Mad2, the latter again in complex with the Mad2 binding region of Mad1 [2]. These results are remarkable for several reasons. The structure of p31^{comet} is strikingly similar to the structure of O-Mad2, implying that p31^{comet} achieves inhibition of the spindle assembly checkpoint by molecular mimickry. p31^{comet} may occupy the O-Mad2 binding site on the C-Mad2–Mad1 receptor and may thereby prevent the formation of C-Mad2–Cdc20 complexes. The structure further shows that the interaction between p31^{comet} and Mad2 depends on residues that are only present in a conformation suitable for binding in C-Mad2, but not in O-Mad2, explaining why p31^{comet} can bind to one and not the other confomer of Mad2. Finally, Yang *et al.* [2] show that p31^{comet} mutants that are defective in C-Mad2 binding are unable to inactivate the spindle assembly checkpoint when expressed in cultured human cells, supporting the notion that p31^{comet} functions by associating with C-Mad2.

These two studies [1,2] are a landmark on our journey towards understanding Mad2 function. In the future, the path of Mad2 research is likely to bifurcate. One direction will have to go even deeper into the inner workings of Mad2 and will need to address how Cdc20 is actually recruited to Mad2 and then locked there inside the C-Mad2 confomer. Does this process occur via formation of the postulated I-Mad2 transition state, and if so, what does this confomer look like and how is its formation catalyzed by the C-Mad2–Mad1 receptor? Answering these questions will not be an easy task, but the impressive recent progress made by the Musacchio and Yu/Luo laboratories awakens the hope that addressing these problems may eventually be feasible. Another research path will have to gravitate away from Mad2, presently the holy grail of the spindle assembly checkpoint field, and explore the steps that follow the interaction of Mad2 with Cdc20. What Mad2 actually does to prevent Cdc20 from activating APC/C is not very clear, nor is it clear how Mad2 interacts with other checkpoint proteins. In cell extracts, Mad2 is found in association with at least two other proteins, BubR1 and Bub3, which are also needed for the function of the spindle assembly checkpoint and which, together with Mad2, assemble into a mitotic checkpoint complex [20]. How Mad2 functions as part of this complex is entirely unclear. Answering these questions will be an exiting task for the future.

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Huddling: Brown Fat, Genomic Imprinting and the Warm Inner Glow

Heat generated by huddling animals is a public good with a private cost and thus vulnerable to exploitation, as illustrated by recent work on rabbits and penguins. Effects of imprinted genes on brown adipose tissue suggest that non-shivering thermogenesis is an arena for intragenomic conflict.

David Haig

"if two lie together, then they have heat; but how can one be warm alone?" (Ecclesiastes 4:11)

Huddling is a widespread cooperative behavior of inactive homeotherm animals. In cold environments, huddling reduces individual heat loss by minimizing

the exposed surface area. Moreover, animals that congregate in an enclosed space can raise the ambient temperature more effectively than a single individual, thus reducing each individual's heat loss per unit of exposed surface. Energy saving through huddling may be substantial. For example, 45 large brown bats (*Eptesicus fuscus*) huddled in a tree cavity expend less than half the energy as they would, were each of them roosting alone [1].

Within a huddle, the fuel consumed by thermogenesis is a direct personal cost to each individual while the benefits are shared by all. Social thermoregulation is, therefore, in principle vulnerable to exploitation by free-riders who skimp on their share of the heating bill. A recent study of emperor penguins reveals the interplay of cooperation and conflict during huddling [2]. Another study by the same group quantifies the benefits of huddling in litters of rabbits [3].

Paternal Endurance and Sib Conclaves

Male emperor penguins endure a four-month fast through the Antarctic winter. During this fast, males live off their fat reserves, incubate a single egg and subsequently feed the newly hatched chick. Males huddle in large groups, especially in bad weather. This allows them to reduce heating costs while maintaining high incubation temperatures. Huddling is essential for successful reproduction because males have insufficient reserves to survive winter on their own [2,4]. Gilbert and colleagues [2] monitored core and subcutaneous body temperatures of five male emperor penguins during their winter fast. Fortuitously (for the researchers), one male lost his egg during a blizzard after 20 days of incubation. The four males who retained their eggs maintained high core temperatures while huddling (36.9 ± 0.3°C). The fifth male, however, reduced his core temperature while huddling (35.5 ± 0.4°C; range 32.8–37.4°C), thereby effectively turning into a net recipient of heat from his warmer neighbors. A plausible interpretation of this observation is that the loss of his egg shifted the marginal costs and benefits of thermogenesis. Once freed from the constraint of providing heat to a developing chick, the male was able to exploit the heat production of other males who still incubated eggs.

Huddling is particularly important for young birds and mammals with high surface area to volume ratios. Huddling behavior has been extensively studied in rat litters [5,6]. Rat pups snuggle closer together at colder temperatures, causing the aggregate surface area of a huddle to expand as temperature rises and contract as temperature falls. During contraction, outer pups attempt to wriggle into the center, whereby individual pups circulate between the surface and center of the pile. When thermogenesis was pharmacologically inhibited in one or more pups, huddles were stable at 15°C if all of the pups or none of the pups were treated, whereas mixed huddles disintegrated [7].

Rabbit mothers, unlike rat dams, suckle their pups once a day for 3–5 minutes, but do not otherwise interact with their offspring. Pups huddle in the absence of their mother and raise their body temperature shortly before her predicted return. Gilbert and colleagues [3] exploited these unusual features of maternal care to assess the benefits of huddling. Rabbit pups were kept at 23–24°C in groups of one, two, four or eight but were returned to their original litters for suckling. In comparison to pups raised alone, pups from groups of eight used 40% less energy between days 3 and 5 and accumulated more fat by day 12. Pups reared alone failed to raise their body temperature before their mother's return and consumed less milk during her brief visits. Thus, energetic savings from huddling were converted into greater access to maternal resources.

Huddling with Relatives

Pups help to reduce each other's heating costs but are also competitors for milk. The evolutionary balance between these two forces will be determined, in part, by the extent of relatedness among huddle-mates. Consider the fate of a hypothetical, newly arisen — and thus rare — allele that reduces thermogenesis in a population in which most individuals contribute to social heating. If huddles consisted of non-relatives, then carriers of the rare allele would be the only members of their huddle to reduce thermogenesis. In this case, a carrier would reduce his own costs of thermogenesis while continuing to receive heat from the rest of the group. If, on the other hand, huddles consisted of full-sibs, then an allele carrier would reduce his heating costs but would also lose heat contributions from half the other members of the huddle who carry a copy of the same allele. Therefore, all other things being equal, alleles for free-loading are more likely to invade a population, and spread to fixation, if huddles consist of non-relatives.

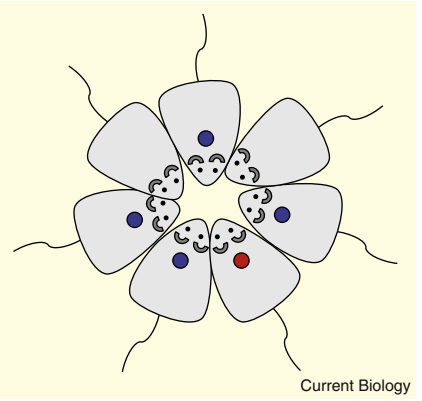

Figure 1. Social heating and genomic imprinting.

Huddling mouse pups from a litter with one mother and multiple fathers. Blue dots represent offspring that have inherited one of the two alleles present in their mother. The red dot represents a paternal allele inherited from one of the fathers. For any particular offspring, its maternal allele is present, on average, in half the other members of the litter but its paternal allele is present in a minority of littermates. Consider a rare allele that reduces thermogenesis: If this is the blue maternal allele, each offspring with the allele will lose heat contributions from three littermates. If this is the red paternal allele, its carrier would continue to be warmed by the other members of the litter. Therefore, maternal alleles are predicted to favor higher levels of thermogenesis than paternal alleles.

Degrees of relatedness can vary among individuals within huddles. In this case, individuals of lower than average relatedness would be expected to exploit the thermogenesis of individuals of higher than average relatedness. Alpine marmots, for instance, hibernate in groups of up to 20 individuals. When the huddle contains juveniles, winter weight loss increases for older sibs of the litter but decreases for less-related individuals [8]. This suggests that older sibs increase thermogenesis to enhance the survival of juveniles, and less-related individuals benefit by reducing their own heat production.

Of particular interest, degrees of relatedness within a huddle may differ for genes of maternal and paternal origin. For example, in a multiple-paternity litter, more individuals will share genes of maternal origin than will share genes of paternal origin. Therefore, maternally expressed imprinted genes are predicted to promote higher contributions to communal heating than the level favored by paternally expressed imprinted genes (Figure 1) [9].

Brown Fat and Genomic Imprinting

Young mammals generate heat by non-shivering thermogenesis in brown adipocytes [10]. At least three imprinted loci influence this process in mice. Two paternally expressed loci, *Pref1/Dlk1* and *Necdin* [11,12], reduce the size of the 'furnace' by inhibiting differentiation of preadipocytes into brown adipocytes [13]. The third imprinted locus, *GNAS*, encodes the G-protein α stimulatory subunit (Gαs) that initiates the cellular events that activate thermogenesis downstream of β-adrenergic receptors [10]. Both maternally and paternally derived *GNAS* alleles produce Gαs in most tissues, but in brown adipose tissue the maternally derived allele is expressed preferentially [14]. By contrast, the paternally derived *GNAS* allele produces the XLαs protein, which antagonizes the effects of Gαs in brown adipose tissue [15]. Thus, *GNAS* produces both a maternally expressed promoter and a paternally expressed inhibitor of non-shivering thermogenesis. This is the pattern that would be predicted if matrilineal relatedness exceeds patrilineal relatedness within huddles. Future studies will test whether this pattern is maintained at other imprinted loci.

The evolution of cooperation has been a major area of theoretical and empirical research in evolutionary biology, but with a perceived need to exploit new study systems for testing theoretical models [16]. Social thermogenesis has certain advantages for studying the stability and breakdown of cooperation. Huddles are spatially localized, and fitness-related variables, such as temperature, body weight or milk consumption, are easily measured. Moreover, pharmacological and genetic interventions are available to adjust how much particular individuals contribute to the collective good.

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Cytosine Methylation: Remaining Faithful

DNA methyltransferase-1 (DNMT1) has a higher specific activity on hemimethylated DNA than on unmethylated DNA, but this preference is too small to explain the faithful mitotic inheritance of genomic methylation patterns. New genetic studies in plants and mammals have identified a novel factor that increases the fidelity of maintenance methylation.

Steen K.T. Ooi and Timothy H. Bestor

In 1975 Art Riggs and Robin Holliday independently predicted the existence of DNA methyltransferases that would methylate only hemimethylated DNA, thereby rendering genomic methylation patterns subject to mitotic inheritance [1,2]. Wigler [3] later showed that methylation patterns were indeed subject to mitotic inheritance, and Groudine and colleagues [4] showed that this inheritance was stable for at least 80 cell doublings in a system that controlled for copy number and integration site effects. Eric Richards and colleagues showed remarkably stable mitotic and meiotic inheritance of CpG methylation patterns in *Arabidopsis thaliana* [5]. Faithful maintenance of methylation patterns is essential for the survival of differentiated cells and may be involved in diseases in which the perpetuation of aberrant DNA-methylation patterns may contribute to disorders of imprinted