Aligning Conservation Priorities Across Taxa in Madagascar with High-Resolution Planning Tools
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Aligning Conservation Priorities Across Taxa in Madagascar with High-Resolution Planning Tools

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Globally, priority areas for biodiversity are relatively well known, yet few detailed plans exist to direct conservation action within them, despite urgent need. Madagascar, like other globally recognized biodiversity hot spots, has complex spatial patterns of endemism that differ among taxonomic groups, creating challenges for the selection of within-country priorities. We show, in an analysis of wide taxonomic and geographic breadth and high spatial resolution, that multitaxonomic rather than single-taxon approaches are critical for identifying areas likely to promote the persistence of most species. Our conservation prioritization, facilitated by newly available techniques, identifies optimal expansion sites for the Madagascar government's current goal of tripling the land area under protection. Our findings further suggest that high-resolution multitaxonomic approaches to prioritization may be necessary to ensure protection for biodiversity in other global hot spots.

Approximately 50% of plant and 71 to 82% of vertebrate species are concentrated in biodiversity hot spots covering only 2.3% of Earth's land surface (*1*). These irreplaceable regions are thus among the highest global priorities for terrestrial conservation; reasonable consensus exists on their importance among various global prioritization schemes that identify areas of both high threat and unique biodiversity (*2*). The spatial patterns of species richness, endemism, and rarity of different taxonomic groups within priority areas, however, rarely align and are less well understood (*3*–*6*). Detailed analysis of these patterns is required to allocate conservation resources most effectively (*7*, *8*).

To date, only a few quantitative, high-resolution, systematic assessments of conservation priorities have been developed within these highly threatened and biodiverse regions (*9*, *10*). This deficiency results from multiple obstacles, including limited data or access to data on species distributions and computational constraints on achieving high-resolution analyses over large geographic areas. We have been able to overcome each of these obstacles for Madagascar, a global conservation priority (*1*, *2*, *11*). Like many other regions (*3*–*6*), Madagascar has complex, often nonconcordant patterns of microendemism among taxa (*12*–*17*), rendering the design of efficient protected-area networks particularly difficult (*4*, *6*). We collated data for endemic species in six major taxonomic groups [ants, butterflies, frogs, geckos, lemurs, and plants (table S1)], using recent robust techniques in species distribution modeling (*18*, *19*) and conservation planning (*20*, *21*) to produce the first quantitative conservation prioritization for a biodiversity hot spot with this combination of taxonomic breadth (2315 species), geographic extent (587,040 km^2), and spatial resolution (30–arc sec grid = ~0.86 km^2).

Currently, an important opportunity exists to influence reserve network design in Madagascar, given the government's commitment, announced at the World Parks Congress in 2003, to triple its existing protected-area network to 10% coverage (*22*). Toward this goal, our high-resolution analysis prioritizes areas by their estimated contribution to the persistence of these 2315 species and identifies regions that optimally complement the existing reserve network in Madagascar.

We input expert-validated distribution models for 829 species and point occurrence data for the remaining species [those with too few occurrences to model, called rare target species (RTS)] into a prioritization algorithm, Zonation (*20*, *21*), which generates a nested ranking of conservation priorities (*23*). Species that experienced a large proportional loss of suitable habitat (range reduction) between the years 1950 and 2000 were given higher weightings [equation 2 of (*23*), (*24*)]. We evaluated all solutions [defined here as the highest-ranked 10% of the landscape to match the target that Madagascar has set for conservation (*22*)] in two ways: (i) percent of species entirely

absent from the solution ["complete gaps" (*11*)] and (ii) proportional representation of species.

Avoiding complete gaps for all species considered, or "minimal representation," is a basic goal of conservation prioritization (*8*) and can be accomplished in only 1020 grid squares (0.1% of the area of Madagascar) in a multitaxon analysis. The single-taxon solutions (fig. S1), however, did a poor job of minimally representing other species (Table 1) because of their low overlap (fig. S2). In single-taxon solutions, 25 to 50% of RTS species from other taxa were entirely omitted (Table 1A). Zero to 18% of modeled species were omitted, depending on whether evaluation was based on actual occurrence points (Table 1B) or distribution models (Table 1D). Overall, the use of any single-taxon solution would result in 16 to 39% of all species ending up as complete gaps (Table 1C, based on actual occurrence records).

In addition to ensuring minimal representation, our goal is to maximize proportional representation (the proportion of distribution or occurrence points) of species, especially those most vulnerable to extinction, in order to increase the probability of their persistence (*11*). In single-taxon solutions, we found that species from other taxa would often be represented at lower levels than the target taxon. Mean proportional representation for modeled species outside of the taxon was lower by a factor of 1.2 to 1.5 relative to the target taxon for all groups except plants (Fig. 1A), which include the most species and the smallest-ranged species within this data set, making it comparatively difficult to protect large proportions of each species even in the plant-specific solution. Similarly, single-taxon solutions contained only 69 to 83%, on average, of the occurrence points for included (species that are represented by at least one record) RTS outside the target taxon, as compared to 100% of RTS records for species within the target taxon (Table 1E). Thus, any conservation prioritization based on a single surrogate taxon would be of limited utility for identifying conservation priorities across taxa in Madagascar.

The ideal solution to the surrogacy problem is to include all species in a single analysis (Fig. 2A), thus avoiding complete gaps (Table 1, last column) while optimizing proportional representation across all taxa. Until now, because of computational constraints, such analyses have not been feasible for this spatial resolution, geographic extent, and number of taxa. Figure S3A shows what can be achieved with the core-area Zonation method when used with weightings that account for historical range reductions. Without this weighting scheme, two species with the same current range size could be included at identical proportional representation, even though one had experienced a precipitous decline in range whereas the other had not. This approach thus prioritizes two classes of vulnerability. Narrow-ranged species, which are vulnerable to habitat loss coincident with their small ranges, are inherently prioritized by the Zonation algorithm [equation S1 of (*23*)]. Species that have suffered extensive recent range reductions (red dots in fig. S3) are additionally prioritized by their weightings, and the proportion of their historical (baseline) range included is thus increased.

Covering all six taxonomic groups simultaneously necessarily invokes tradeoffs, decreasing, for example, the proportions of species distributions represented in each taxon significantly relative to its own single-taxon solution (Fig. 1B, –0.04 ± 0.002 SE, paired Wilcoxon signed-ranks test, $P < 0.0001$). To assess this tradeoff, we calculated a potential extinction risk for modeled species based on future distributional loss under the single- and multitaxon solutions, assuming loss of all habitat outside of prioritized areas and an aggregate species-area response (*24*). The increase in potential extinction risk for each taxonomic group incurred under the multitaxon solution relative to its own (fig. S4) constitutes the cost of including hundreds of species in the protected-area network that would otherwise be omitted (Table 1C).

We compared our multitaxon solution (Fig. 2A) against the actual parks selected during the recent protected-area expansion phase of 2002–2006 that has increased the total reserve coverage from 2.9 to 6.3% of Madagascar (Fig. 2B). The mean proportion of modeled species distributions included in the multitaxon solution (using the top 6.3% prioritized to compare with the area protected by 2006) was not significantly higher than in the actual selections (+0.004 ± 0.002 SE, paired test, NS), as is expected because of tradeoffs among species (that is, given the fixed area of 6.3%, some species increased in representation when the optimized solution was compared to the actual solution, whereas others necessarily decreased, resulting in no mean change). The multitaxon solution, however, included all species, whereas the actual selections entirely omitted 28% of species (based on actual occurrence points, fig. S5). In addition, proportions included for the species with narrowest ranges or largest scores for the proportional range-reduction index were significantly larger in the multitaxon solution (at 6.3% of area) as compared to the actual selection [Kolmogorov-Smirnov two-sample test, first (smallest) quartile of range size, $D = 0.28$, $n = 207$ species, $P < 0.001$; fourth (largest) quartile of proportional range-reduction index, $D = 0.149$, $n = 207$ species, $P = 0.001$].

Finally, because we are operating in a real-world conservation context and many protected areas have already been established in Madagascar, we developed a realistic Zonation solution,

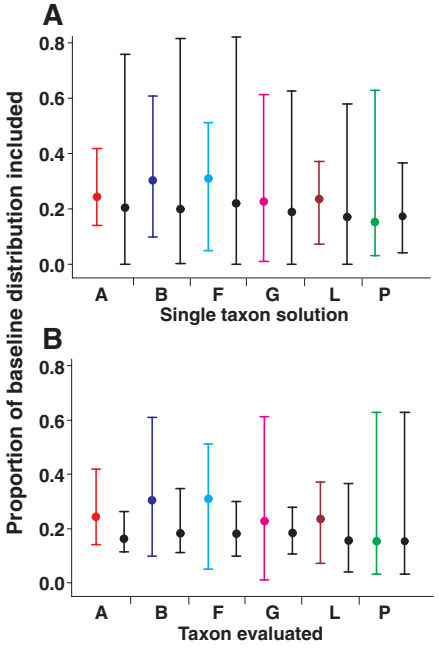

Fig. 1. Evaluating the top 10% of Zonation solutions for single- and multitaxon solutions. (**A**) The minimum, mean, and maximum proportion of the baseline (1950) distribution included for each taxonomic group [red, ants (A); blue, butterflies (B); cyan, frogs (F); pink, geckos (G); brown, lemurs (L); green, plants (P)] in its taxon-specific solution at 10% (fig. S1, A to F), compared to the corresponding mean and range for all other taxa (not including the solution taxon) if this particular single-taxon solution were to be adopted (black). (**B**) The minimum, mean, and maximum proportion of the baseline distribution for each taxonomic group [colors and labels as in (A)] under its own individual solution (maps in fig. S1, A to F), compared to the values obtained for its taxonomic group only under the multitaxon solution (black, map in Fig. 2A).

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optimized to expand on existing protected areas (6.3%) by adding an additional 3.7% of area (Fig. 2B, constrained solution). Like the unconstrained solution (Fig. 2A and Table 1), this solution (Fig. 2B) omits no species. The proposed expansion achieves relatively large increases in mean proportional representation (+0.05 ± 0.001 SE of modeled species' distributions and +58.8 ± 1.1% SE of RTS' occurrences). Most important, it realizes gains among the most vulnerable species, because of both the algorithm (*20*, *21*) and the weighting system used. Among modeled species, those that have already lost much of their range (Fig. 3, A to C; red indicates the highest quartile of proportional range-reduction index) or are currently narrow-ranged (Fig. 3, D to F; red indicates the smallest quartile of range) increase most in proportional representation when moving from current parks (Fig. 3, B and E) to the constrained optimized solution (Fig. 3, C and F). For RTS species, expansion from current parks to the optimized solution would increase mean proportional representation to 99.9 ± 0.1% SE of occurrences from 0% for gap species (39% of all RTS, fig. S5) or 67.8 ± 1.9% SE for included species (fig. S6). Thus, although the protected areas selected to date have captured a relatively high proportion of Madagascar's species (~70% of species considered here, fig. S5), careful selection of the remaining 3.7% of area (as in the plan proposed in Fig. 2B) can produce further substantial conservation gains, both by including many more species and by increasing the proportional representation of the most vulnerable ones.

Our analysis provides fresh insights into conservation needs for Madagascar, identifying, for example, several regions within the central plateau massifs and littoral forests as priorities (Fig. 2): areas with relatively low forest cover but considerable endemism that have been historically neglected in favor of protecting large forest blocks. Although our national-scale analysis identifies important biodiversity priorities at high resolution, precise delineation of protected areas requires taking socioeconomic factors into account (*25*). Within these priority areas, those that are most vulnerable to habitat destruction or are most highly ranked (fig. S7) should receive immediate attention (*26*). Although conservation areas must be identified by the end of 2008, final refinement and legal designation will not be completed until 2012. Thus, time is available for implementation of an iterative process (*8*): rerunning this analysis to select optimal replacement sites each time areas within the solution are definitively rejected or destroyed, or alternate areas are definitively selected. Such updates could

Table 1. Surrogacy of higher taxa, comparing single- and multitaxon solutions. Section A, percentage of complete gap species for RTS species (n = 1486). B, percentage of complete gap species for modeled species (n = 829). C, percentage of complete gap species for all species (modeled and RTS, n = 2315). Sections A, B, and C are based on occurrence data, and complete gaps are species with no points included in the solution. The diagonals and the multitaxon columns have no unrepresented species, demonstrating as expected that Zonation includes all species considered within its solution. For D, the gap analysis was performed with models rather than occurrence points. E, mean percent of occurrence points included for nongap RTS species (species represented by at least one point in the solution). n.a., not applicable because all species are included in the solution by definition.

		Taxon targeted by zonation solution						
	Taxon assessed	Ants	Butterflies	Frogs	Geckos	Lemurs	Plants	All taxa
A. Percent of unmodeled (RTS) species unrepresented, based on point occurrence records	Ants	0	21.3	28.9	33.6	32.4	26.9	0
	Butterflies	14.5	0	22.1	25.2	38.9	24.4	0
	Frogs	34.1	25.7	0	30.7	25.7	21.2	0
	Geckos	26.9	23.1	23.1	0	26.9	19.2	0
	Lemurs	42.9	50.0	50.0	71.4	0	35.7	0
	Plants	45.2	52.3	42.8	62.2	54.8	0	0
	All species except target taxon	40.0	42.4	37.7	50.2	45.5	24.5	n.a.
B. Percent of modeled species unrepresented, based on point occurrence records	Ants	0	0	5.5	2.7	0	0	0
	Butterflies	0	0	4.7	0.6	0	0.6	0
	Frogs	5.0	5.0	0	5.0	0	0	0
	Geckos	0	0	0	0	0	0	0
	Lemurs	3.2	6.5	3.2	9.7	0	0	0
	Plants	13.3	14.1	23.4	26.2	16.4	0	0
	All species except target taxon	9.3	11.4	16.4	17.5	10.5	0.3	n.a.
C. Percent of modeled and RTS unrepresented, based on point occurrence records	All species except target taxon	28.3	32.3	29.6	38.5	33.2	16.2	0
D. Percent of modeled species with no part of their model protected by the Zonation solution	Ants	0	0	0	0	0	0	0
	Butterflies	0	0	1.2	0	0	0	0
	Frogs	0	0	0	0	0	0	0
	Geckos	0	0	0	0	0	0	0
	Lemurs	0	0	3.2	0	0	0	0
	Plants	1.6	0.4	8.0	2.0	1.6	0	0
	All species except target taxon	1.1	0.3	5.4	1.2	1.0	0.0	n.a.
E. Mean percent point occurrence records included for (nongap) RTS species only	Ants	100.0	84.9	87.6	80.5	75.7	77.1	100.0
	Butterflies	77.4	100.0	84.3	81.0	68.9	70.1	100.0
	Frogs	71.8	75.7	100.0	75.4	76.2	75.5	100.0
	Geckos	75.7	73.7	74.8	100.0	64.3	69.9	100.0
	Lemurs	68.1	49.9	45.6	39.0	100.0	56.3	100.0
	Plants	65.7	66.5	71.6	65.9	61.1	99.9	99.86
	All species except target taxon	68.7	72.8	76.6	72.7	67.5	74.4	n.a.

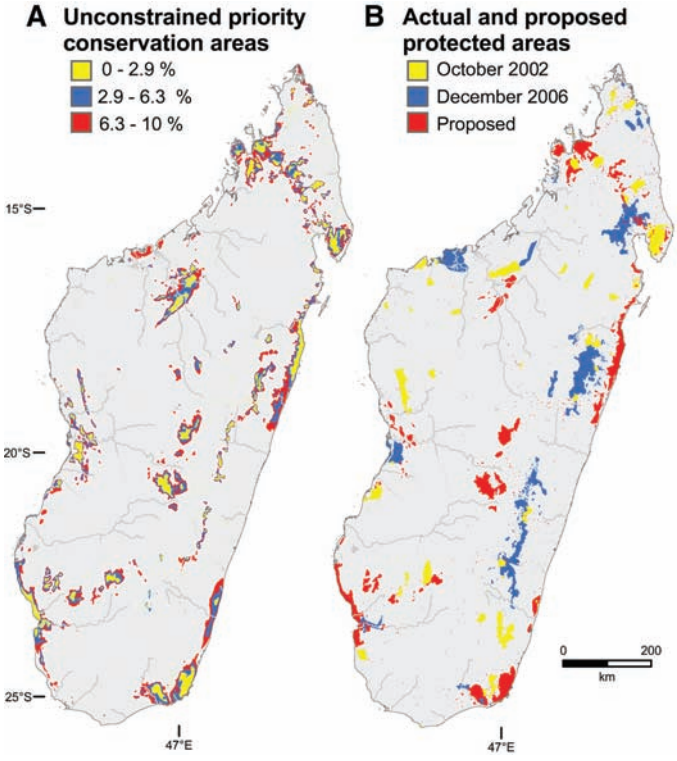

Fig. 2. Conservation priority zones in Madagascar. (**A**) Unconstrained multitaxon solution, showing what would have been selected based on these 2315 species if no areas were already protected. Colors indicate priority level: The top-ranked 2.9% priority areas are shaded yellow (equivalent to the area actually protected by 2002), the next-ranked priorities to 6.3% are blue (equivalent to the area actually protected by 2006), and the next-ranked priorities to 10% (equivalent to the conservation target) are red. (**B**) Constrained multitaxon solution, expanding (red) from existing parks in 2006 (yellow + blue = 6.3% of area) to 10% protection. The red areas are thus those that our analysis selects as the most important areas to consider for expansion of the current reserve network.

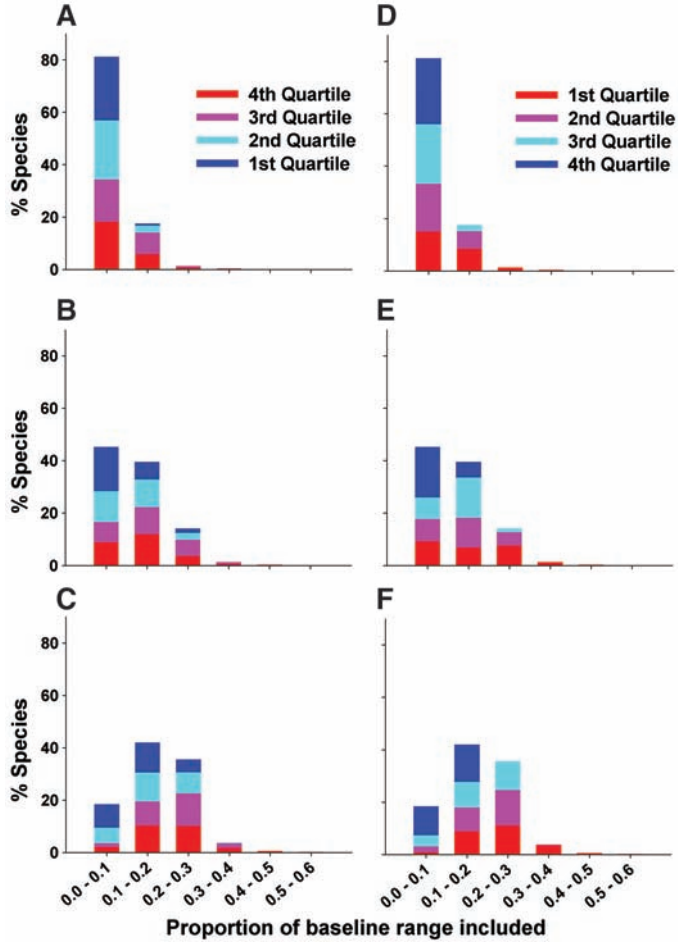

Fig. 3. Proportions of baseline (1950) species ranges (modeled) included at different phases of park expansion, as frequency histograms. (**A** to **C**) Within each histogram, species are coded by their proportional range-reduction index (weights used in Zonation), binned by quartiles, with the fourth quartile (red) representing the largest reductions. (**D** to **F**) Within each histogram, species are coded by their current range size, binned by quartiles, with the first quartile (red) representing the smallest-ranged species. [(A) and (D)] Protected areas designated by the year 2002, equaling 2.3% of the landscape (shaded yellow in Fig. 2B). [(B) and (E)] Protected areas designated by the year 2006, 6.3% of the landscape (shaded yellow and blue in Fig. 2B). [(C) and (F)] Constrained optimized expansion to 10% of the landscape (shaded yellow, blue, and red in Fig. 2B).

incorporate other taxonomic groups, new species records, and changing species designations (*27*). Our results suggest that conducting comparable analyses for other globally biodiverse areas is not only feasible but necessary, because of the inadequacy of single-taxon analyses to identify cross-taxon priorities and the need to develop high-resolution priorities within hot spots. As conservation targets are approached, optimization techniques become particularly critical to guide the final, toughest choices, so as to increase both the future representation of species in reserves and the probability that populations of these species will persist.

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evaluated species distribution models and Zonation solutions for their taxa. A.R. conducted the geographic information system (GIS) analyses to produce the SAPM priority map (Fig. 2B, black outlines). C.K. and A.C. wrote the initial draft of the manuscript; all authors commented on subsequent drafts.

Supporting Online Material
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An Agonist of Toll-Like Receptor 5 Has Radioprotective Activity in Mouse and Primate Models

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The toxicity of ionizing radiation is associated with massive apoptosis in radiosensitive organs. Here, we investigate whether a drug that activates a signaling mechanism used by tumor cells to suppress apoptosis can protect healthy cells from the harmful effects of radiation. We studied CBLB502, a polypeptide drug derived from *Salmonella* flagellin that binds to Toll-like receptor 5 (TLR5) and activates nuclear factor–κB signaling. A single injection of CBLB502 before lethal total-body irradiation protected mice from both gastrointestinal and hematopoietic acute radiation syndromes and resulted in improved survival. CBLB502 injected after irradiation also enhanced survival, but at lower radiation doses. It is noteworthy that the drug did not decrease tumor radiosensitivity in mouse models. CBLB502 also showed radioprotective activity in lethally irradiated rhesus monkeys. Thus, TLR5 agonists could potentially improve the therapeutic index of cancer radiotherapy and serve as biological protectants in radiation emergencies.

The toxicity of high-dose ionizing radiation (IR) is associated with induction of acute radiation syndromes (*1*) involving the hematopoietic system (HP) and gastrointestinal tract (GI). The extreme sensitivity of HP and GI cells to genotoxic stress largely determines the adverse side effects of anticancer radiation therapy and chemotherapy (*2*). Development of radioprotectants for medical and biodefense applications has primarily focused on antioxidants that protect tissues (*3*) and cytokines that stimulate tissue regeneration (*4*).

Here, we have explored whether radioprotection can be achieved through suppression of apoptosis, the major mechanism underlying massive cell loss in radiosensitive tissues (*5*–*7*). Specifically, we have attempted to pharmacologically mimic an antiapoptotic mechanism frequently acquired by tumor cells, i.e., constitutive activation of the nuclear factor–κB (NF-κB) pathway (*8*). NF-κB is a transcription factor that plays a key role in cellular and organismal response to infectious agents as a mediator of innate and adaptive immune reactions. The link between NF-κB and the mammalian response to IR has been established by previous work showing that GI radiosensitivity is enhanced in mice with a genetic defect in NF-κB signaling (*9*). Activation of NF-κB induces multiple factors that contribute to cell protection and promote tissue regeneration, including apoptosis inhibitors, reactive oxygen species scavengers, and cytokines. Finally, NF-κB activation is among the mechanisms by which tumors inhibit function of the p53 tumor suppressor pathway (*10*), one of the major determinants of radiosensitivity (*11*).

In order to activate NF-κB in GI cells without inducing acute inflammatory responses, we studied factors produced by benign microorganisms in the human gut that activate NF-κB by binding to Toll-like receptors (TLRs) expressed by host cells (*12*). Stimulation of TLR signaling by commensal microflora plays a protective role in the GI tract (*13*). In particular, we focused on TLR5, which is expressed on enterocytes, dendritic cells (*14*), and endothelial cells of the small intestine lamina propria (*15*). Endothelial cell apoptosis has been identified as an important contributor to the pathogenesis of GI acute radiation syndrome (*16*). The only known ligand and agonist of TLR5 is the bacterial protein flagellin (*17*).

To investigate whether flagellin has in vivo radioprotective activity, we injected flagellin purified from *Salmonella enterica* serovar Dublin (*18*) into NIH-Swiss mice 30 min before total-body γ irradiation (TBI). Treatment with 0.2 mg/kg of body weight of flagellin protected mice from lethal doses of 10 and 13 Gy that induce mortality from HP and GI acute radiation syndromes, respectively (Fig. 1A). Flagellin did not rescue mice from 17 Gy TBI but prolonged their median survival from 7 to 12 days. The dose-modifying factor (DMF, the fold change in irradiation dose lethal for 50% of animals) of CBLB502 in NIH-Swiss mice was 1.6, exceeding that of other radioprotective compounds, such as cytokines or amifostine, used at nontoxic doses (*3*).

To reduce the immunogenicity and toxicity of flagellin, we took advantage of studies that mapped the TLR5-activating domains of flagellin to its evolutionarily conserved N and C termini (Fig. 1B) (*19*). We tested a series of engineered flagellin derivatives for NF-κB activation in vitro (Fig. 1B and fig. S1). The most potent NF-κB activator, designated CBLB502, included the complete N- and C-terminal domains of flagellin separated by a flexible linker (fig. S1). CBLB502 produced in *Escherichia coli* as a recombinant protein retains entirely the NF-κB–inducing activity and exceptional stability of flagellin (*18*), yet is substantially less immunogenic (fig. S2). It is also less toxic than flagellin, with a maximum tolerated dose (MTD) in mice of 25 mg/kg as compared with the 12 mg/kg MTD of flagellin (*20*). Flagellin derivatives that failed to activate NF-κB in vitro did not provide radioprotection in vivo (one example is shown in Fig. 1C), which suggested that activation of TLR5 signaling is necessary for radioprotection.

To test whether CBLB502 retained the radioprotective efficacy of flagellin, we administered a single injection of the compound (0.2 mg/kg) to NIH-Swiss mice 30 min before 13 Gy TBI. The treatment (*18*) rescued more than 87% of mice from radiation-induced death (Fig. 1C). At this radiation dose, the most powerful previously described radioprotectants provided about 54% protection [amifostine (*21*)] or had no protective effect at all [5-androstenediol (5-AED) or Neumune (*22*)] (Fig. 1C). Notably, the moderate protective effect observed with amifostine against 13 Gy TBI required injection of a dose (150 mg/kg) close to its MTD (200 mg/kg in NIH-Swiss mice). CBLB502 showed a significantly stronger protective effect ($P < 0.05$) when it was injected at less than 1% of its MTD.

To address the practicality of CBLB502 as an antiradiation drug, we investigated the time frame for effective administration of the compound at different radiation doses. CBLB502 protected mice against the very high doses of radiation that induce lethal HP or combined HP and GI syndromes (10 Gy and 13 Gy, respectively) only when injected 15 to 60 min before TBI (Fig. 1D). The compound provided no survival benefit if injected before this time interval or after irradiation.

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