

Supplementary Material for: Cultural Evolution of Central African hunter-gatherers reflects a deep history of interconnectivity

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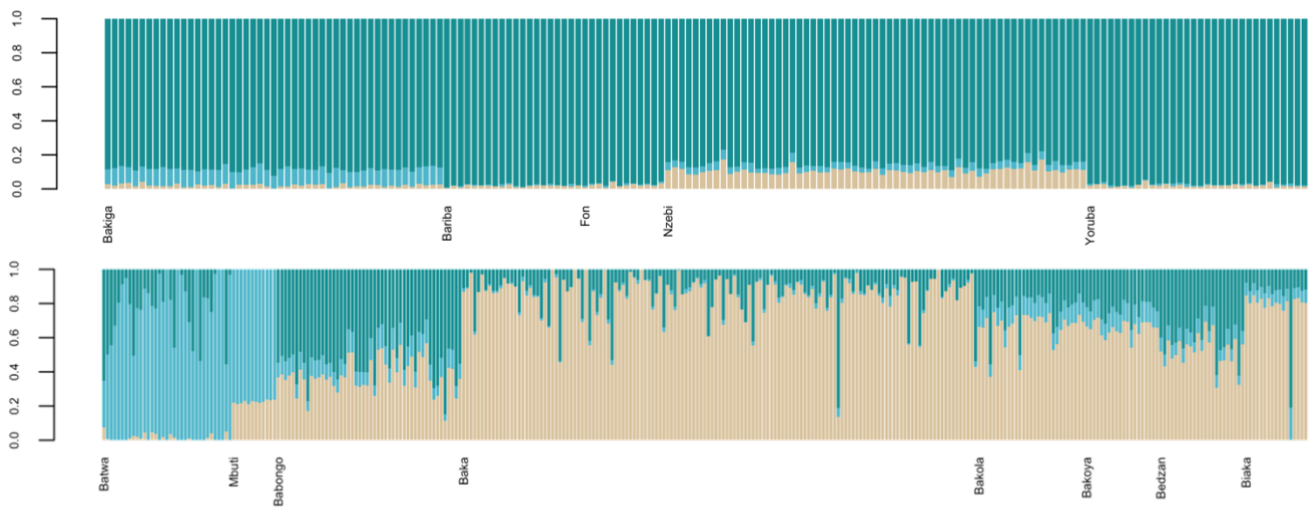


Figure 1. Results from the programme ADMIXTURE on our reduced pruned SNP dataset. Each bar corresponds to an individual and colours represent the proportion of inferred ancestry components from $K=3$ ancestral populations.

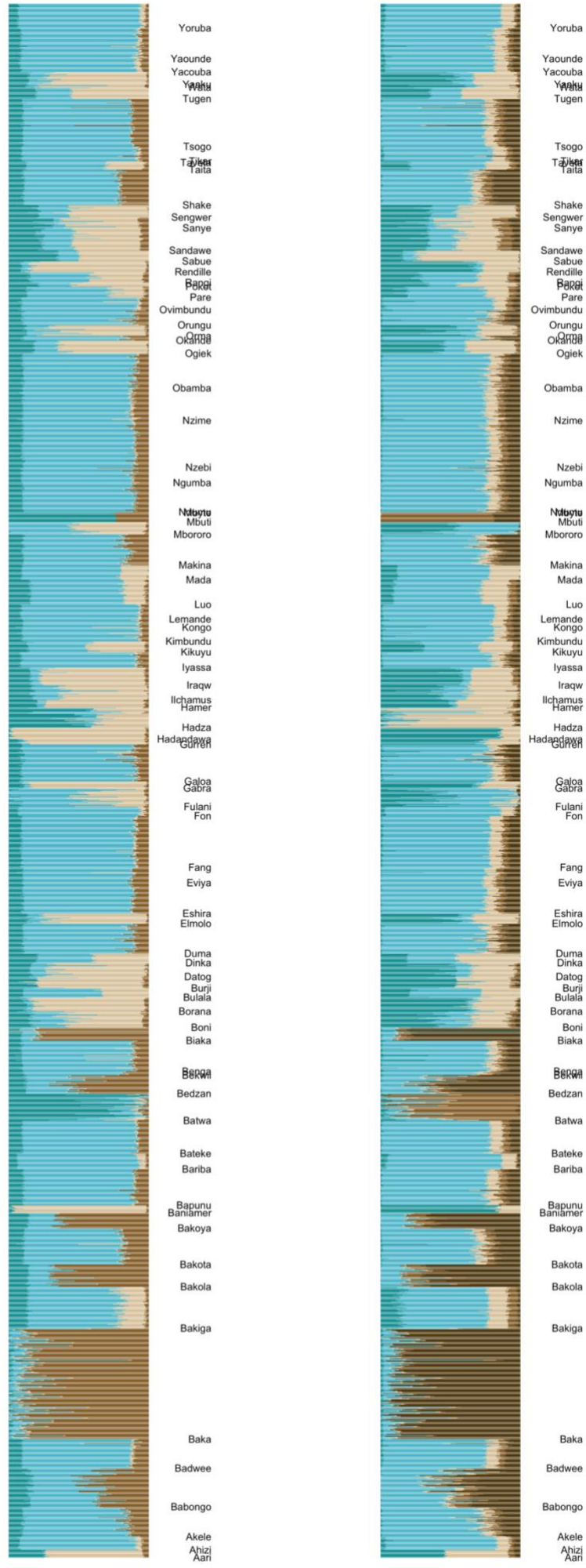


Figure 2. Admixture results on full pruned dataset at $K=4$ (left) and $K=5$ (right)

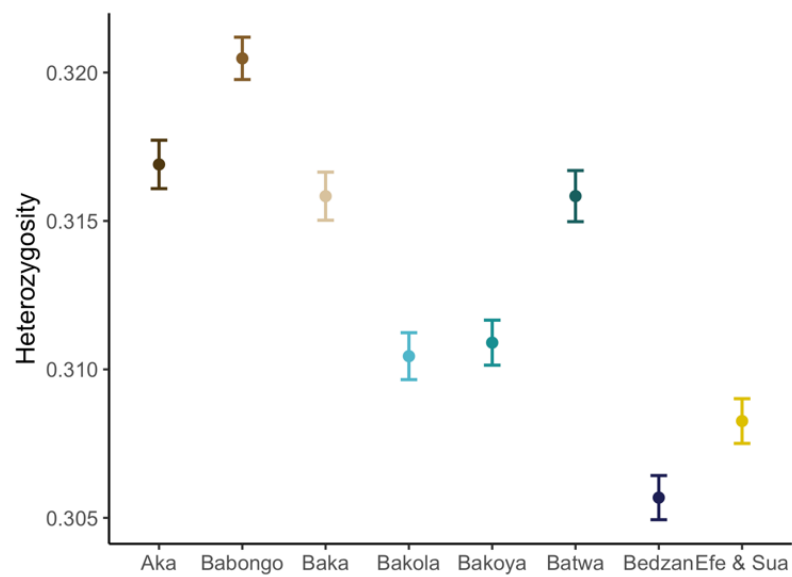


Figure 3. Average heterozygosity proportion of CAHG ancestry components in the populations in our sample.

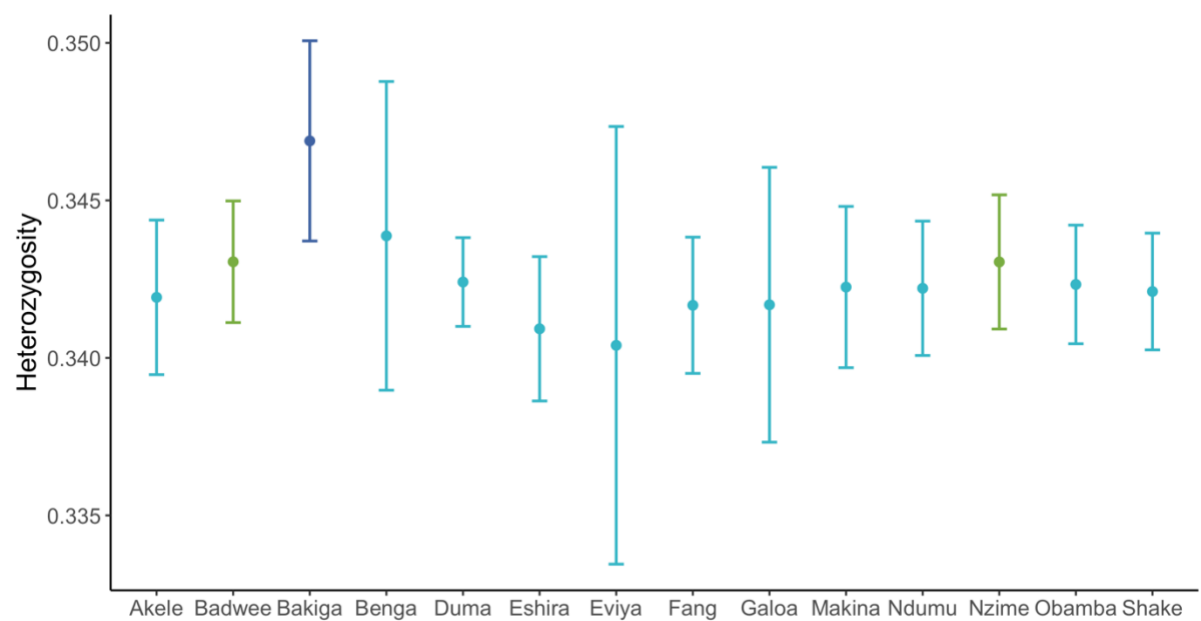


Figure 4. Average heterozygosity of farmer populations neighbouring CAHG in our sample. Dark blue designates Ugandan population, light blue Gabonese populations and green Cameroonian populations.

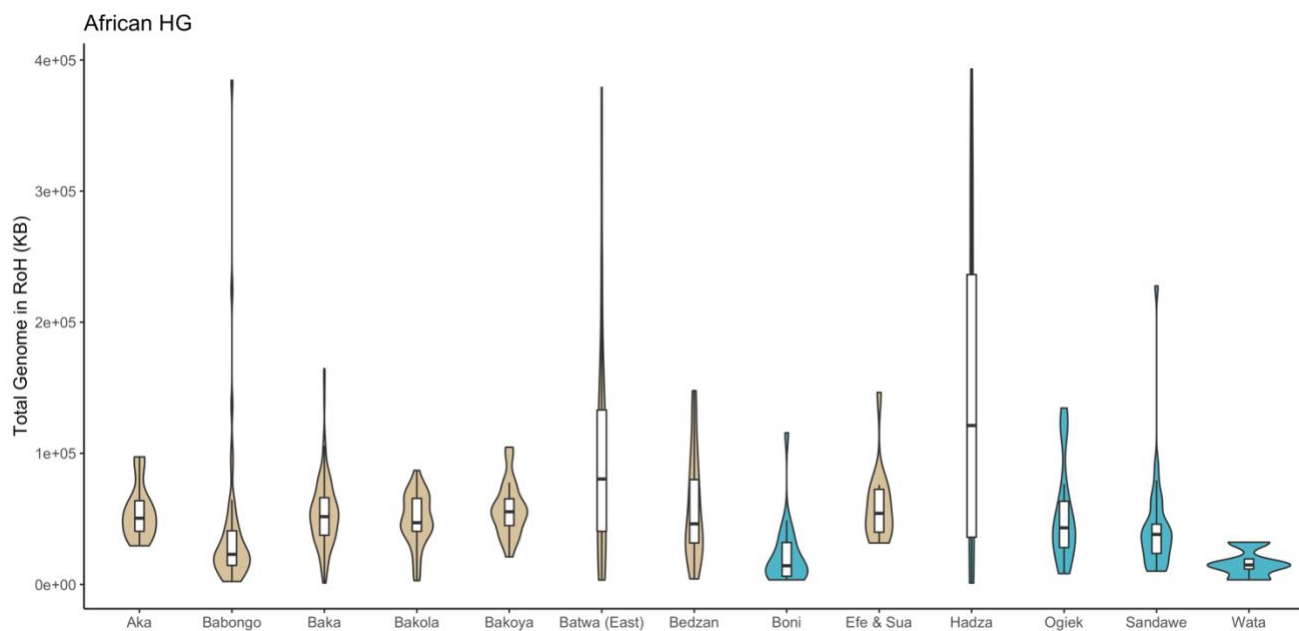


Figure 5. Average length of the genome in runs of homozygosity (RoH) in all African hunter-gatherer populations included in our sample

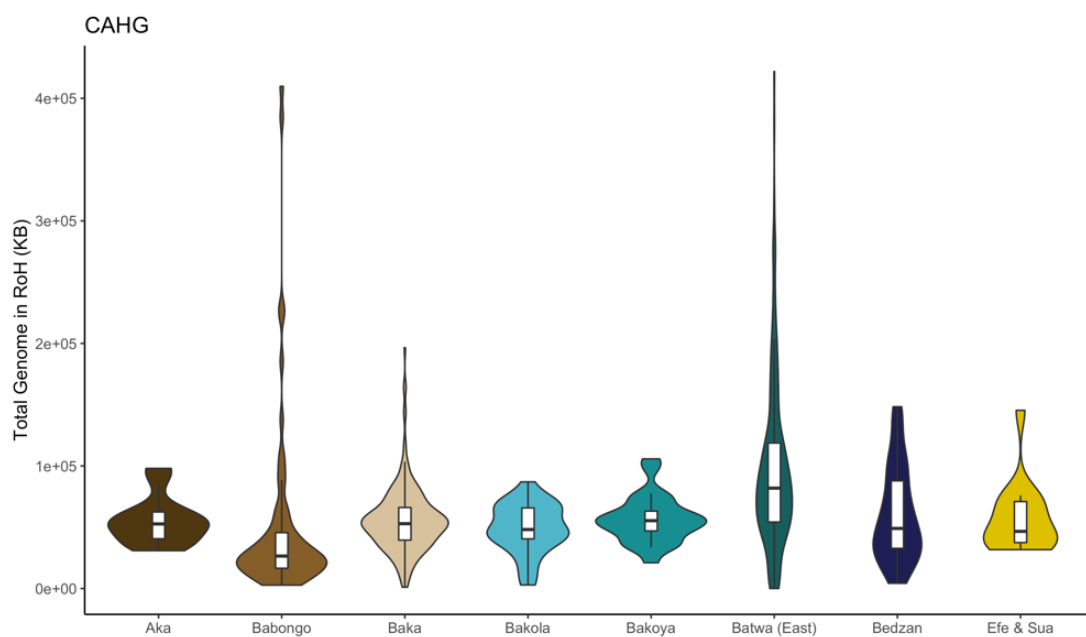


Figure 6. Average length of genome in runs of homozygosity for CAHG populations in our sample

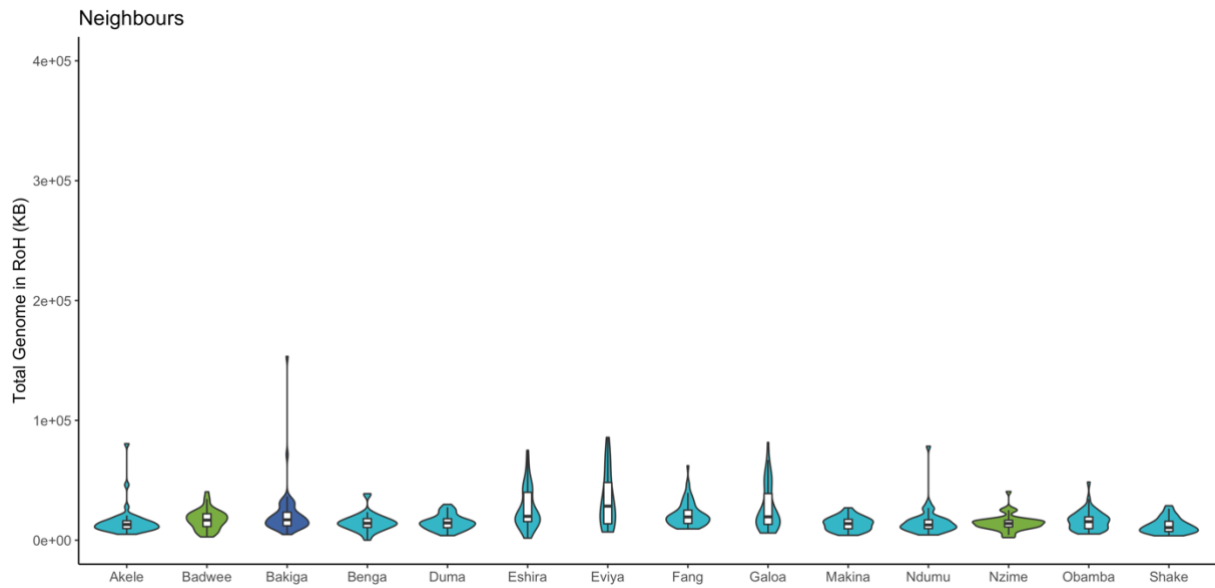


Figure 7. Average length of the genome in runs of homozygosity (RoH) in neighbours of CAHG populations included in our sample.

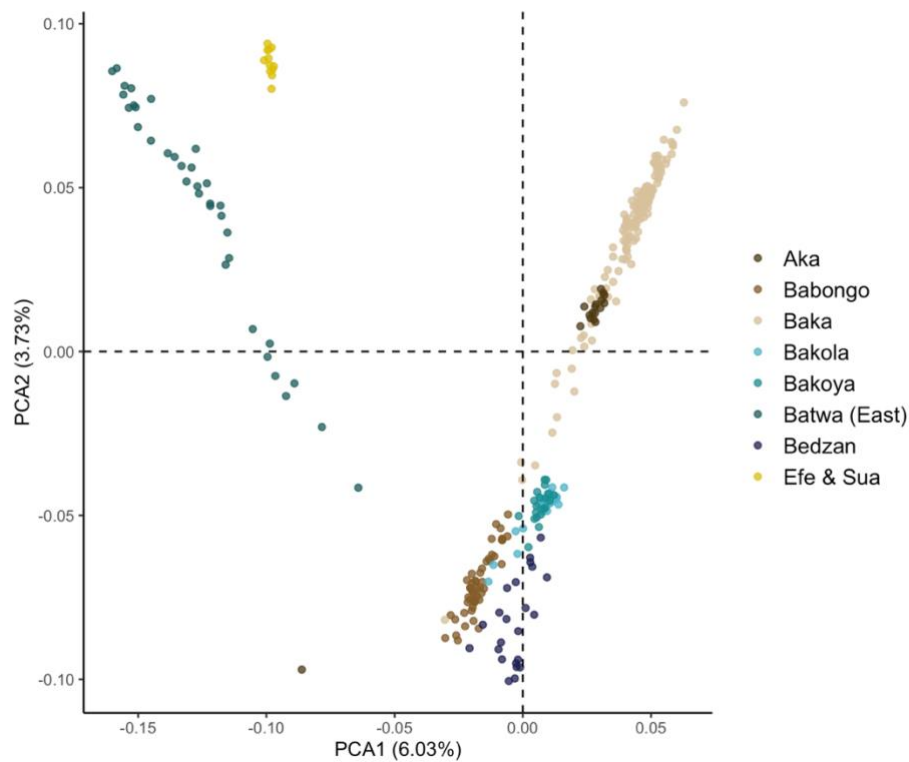


Figure 8. PCA of CAHG with available genome-wide SNP data using the full (unmasked) dataset

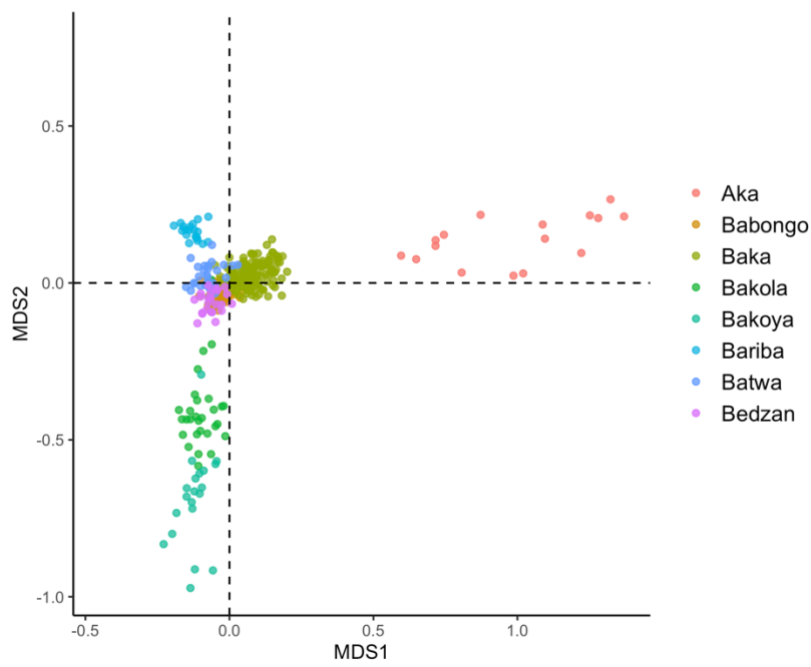


Figure 9. PCA of CAHG with available genome-wide SNP data using exclusively Bantu-associated genomic segments. Note that the Mbuti are not included as they do not carry a sufficient amount of Bantu-associated DNA.

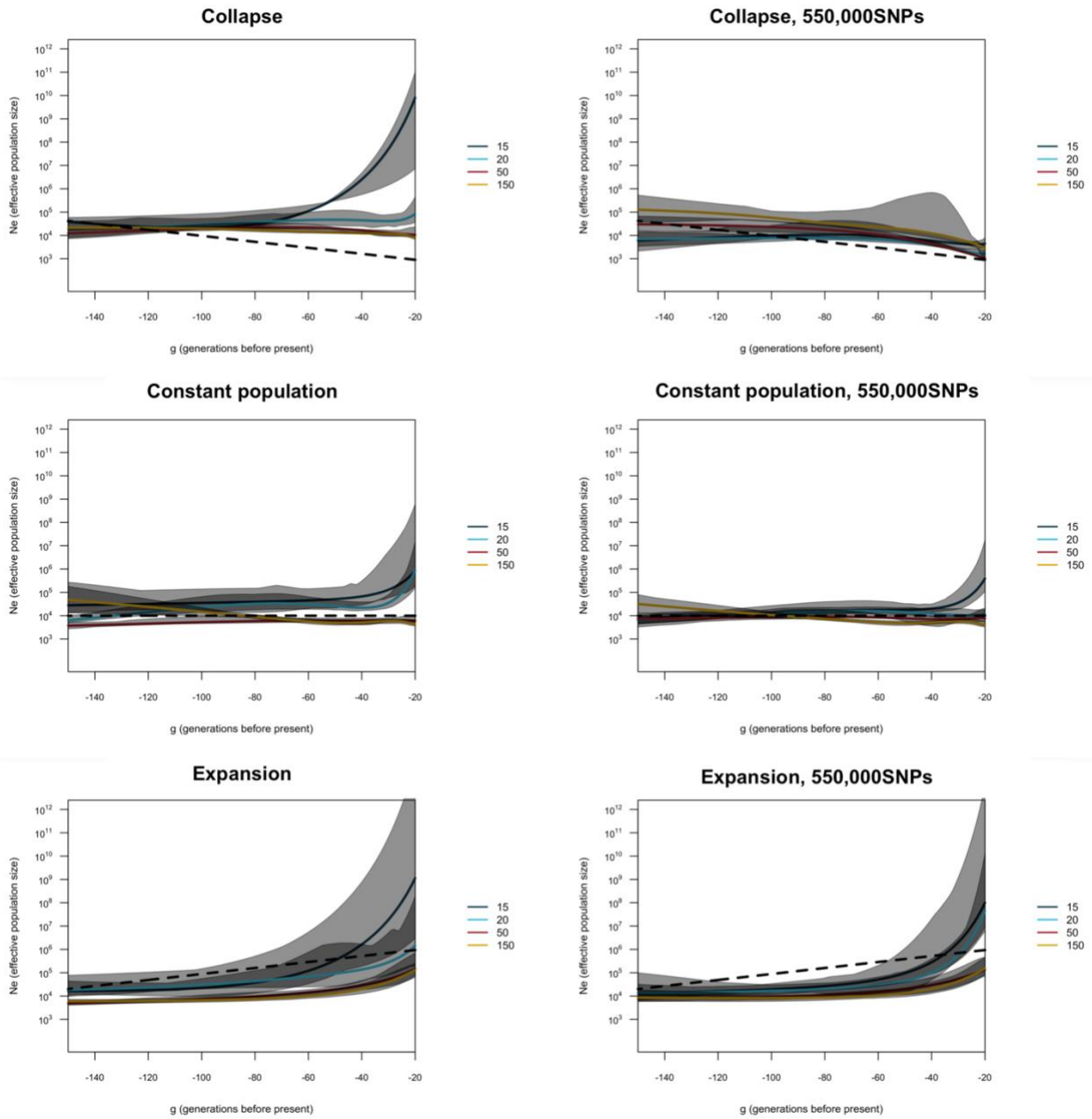


Figure 10. Population size (N_e) estimates from 4,500 years B.P until 600 years B.P inferred from simulated data. Left column shows results from simulated sequence data filtered for alleles with a frequency $<10\%$ and right column from simulated SNP array data. Top row shows results from a declining effective population of initially 45,000 individuals declining at a rate of -0.03 , middle row from a constant effective population of 10,000 individuals and bottom row from a growing effective population of initially 11,000 individuals at a rate of 0.03 . Coloured lines show N_e for simulations containing sample sizes of 15, 20, 50 and 150 individuals. Shaded regions show bootstrap 95% confidence intervals around such estimates and dashed lines show real simulated N_e .

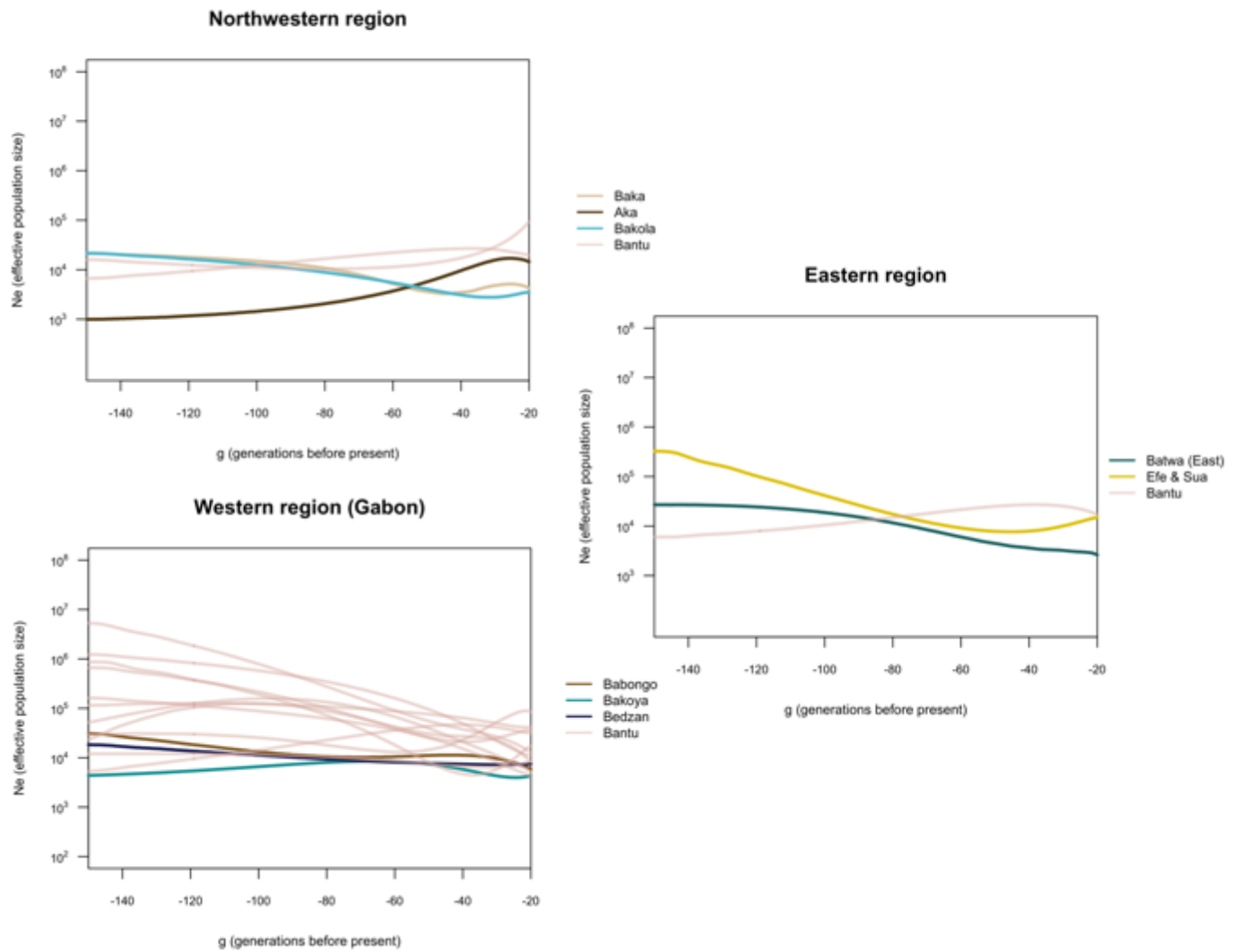


Figure 11. Effective population size (N_e) estimates for CAHG and Bantu-speaking populations inhabiting the same regions from 4,500 years B.P. until 600 years B.P. (A) N_e estimates for northwestern groups include Baka, Aka and Bakola (CAHG) and Bantu-speaking Badwee and Nzime from Cameroon and the Central African Republic. (B) N_e estimates for Eastern groups Mbuti and Eastern Batwa (CAHG) and Bantu-speaking population Bakiga from the Eastern part of the Democratic Republic of Congo and Uganda. (C) N_e estimates Babongo, Bakoya and Bedzan (CAHG) and Bantu-speaking Akele, Benga, Duma, Eshira, Eviya, Fang, Galoa, Makina, Ndumu, Obamba and Shake from Gabon. Each generation represents 30 years.

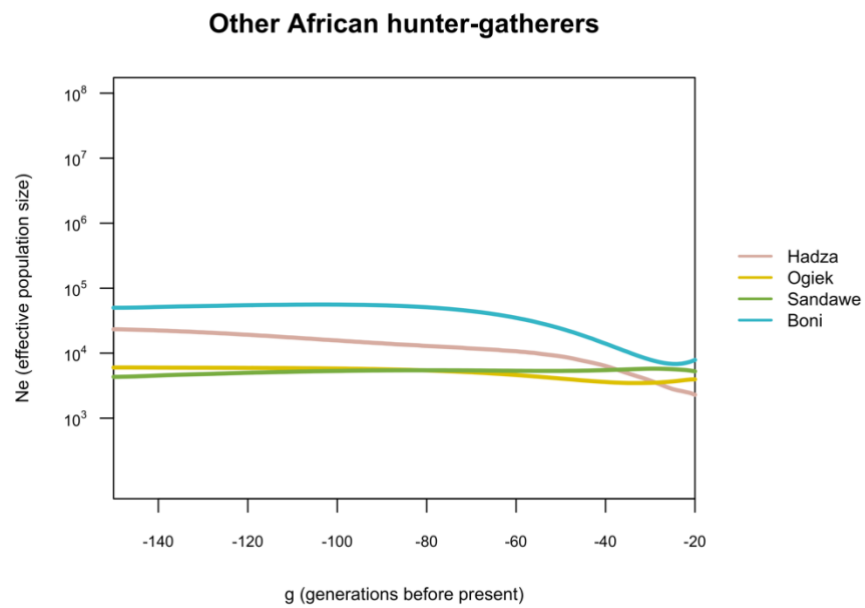


Figure 12. IBDNe results from hunter-gatherer populations from Eastern Africa included in our sample

Masked Fst

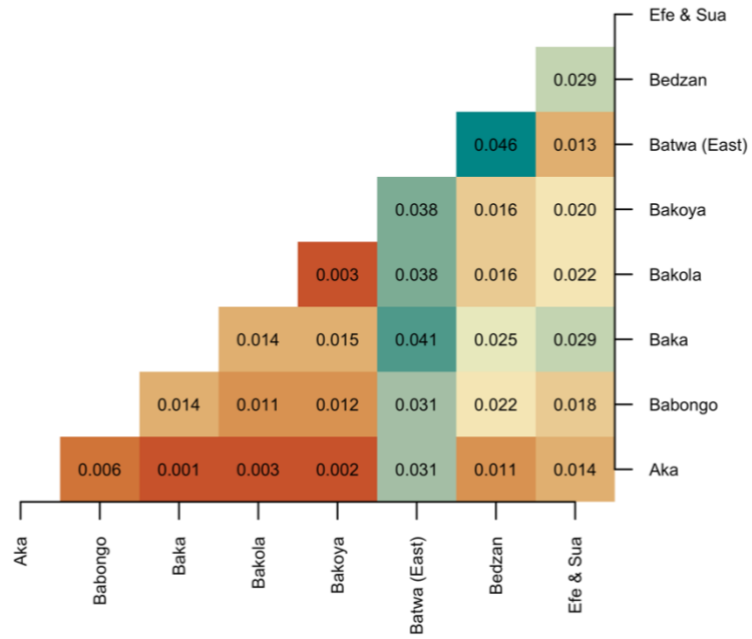


Figure 13. Inter-population F_{ST} using only CAHG ancestry components

Masked Fst, no IBD segments

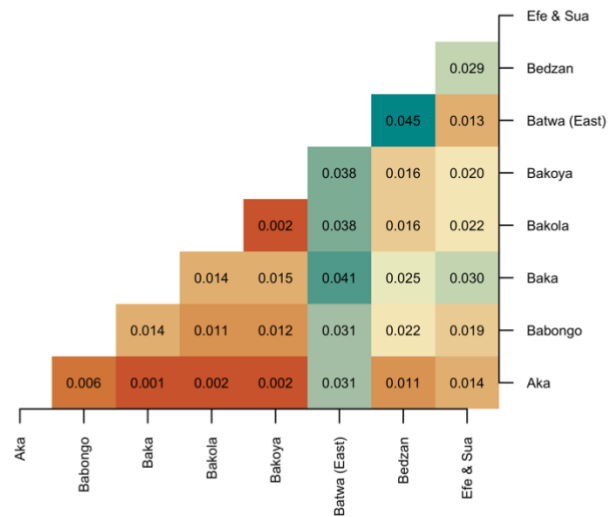


Figure 14. Pairwise F_{st} matrix using only CAHG ancestry components and excluding shared IBD segments between populations

Masked Fst, only Bantu segments

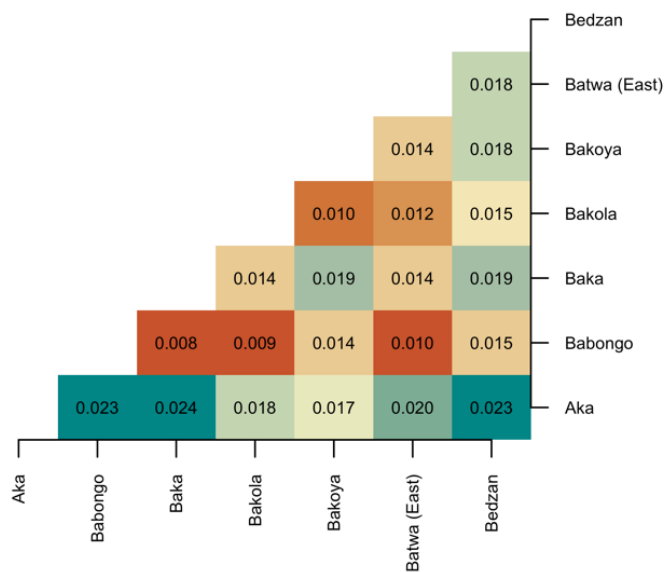


Figure 15. Pairwise F_{st} matrix using only Bantu ancestry components

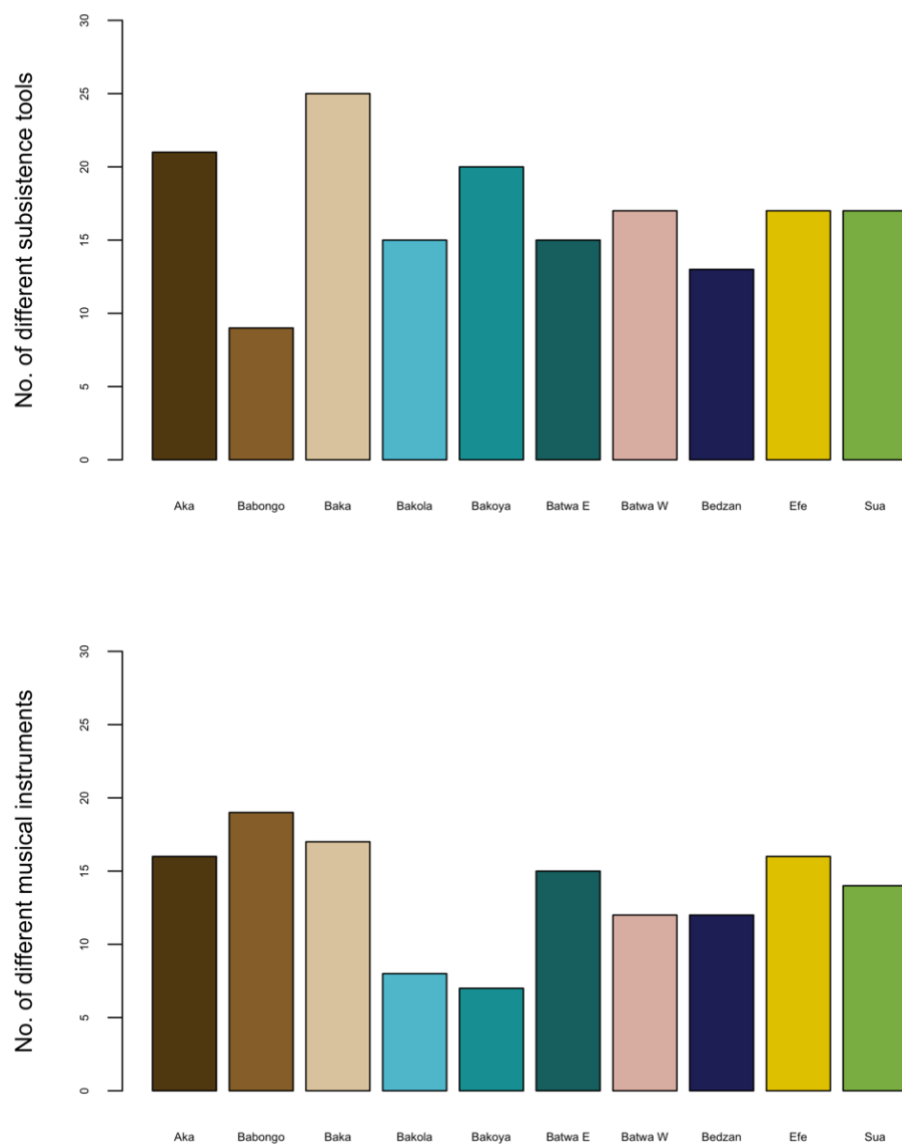


Figure 16. Count of different subsistence tools (top) and musical instruments (bottom) for the 10 CAHG populations in our sample.

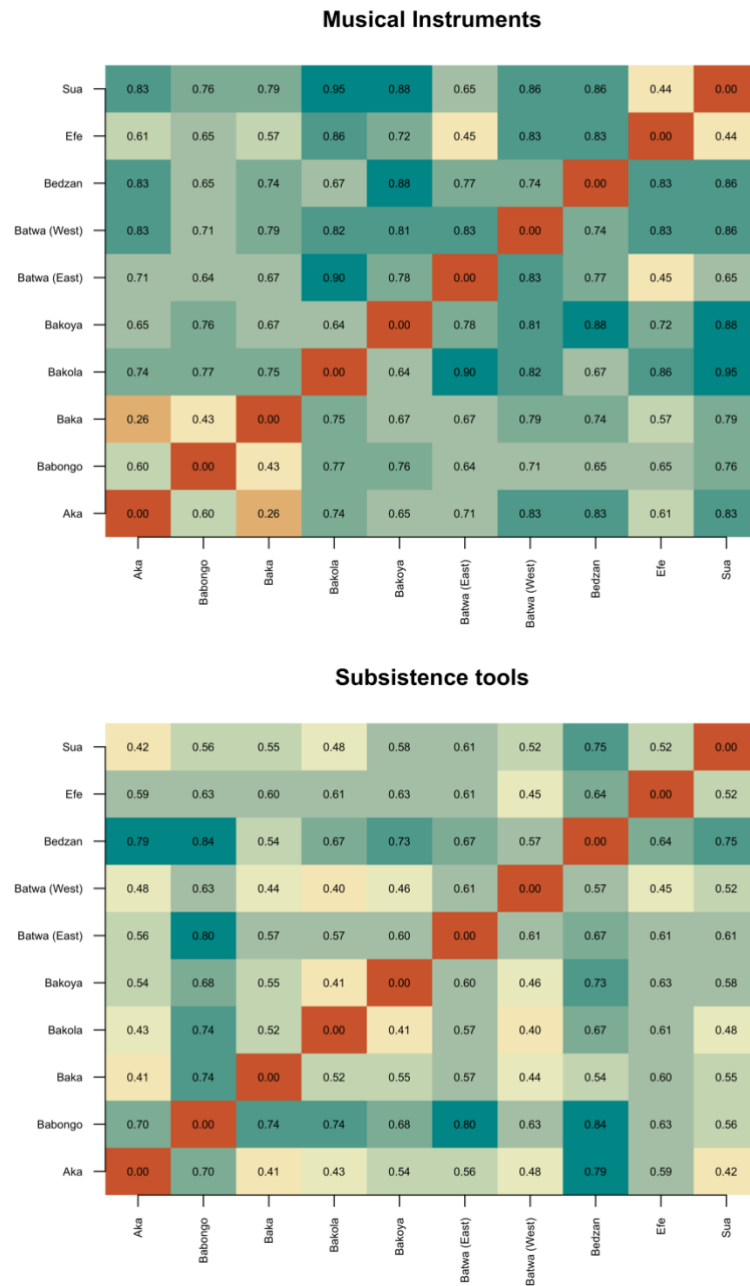


Figure 17. Pairwise Jaccard distances in repertoires of musical instruments and foraging-related toolkits

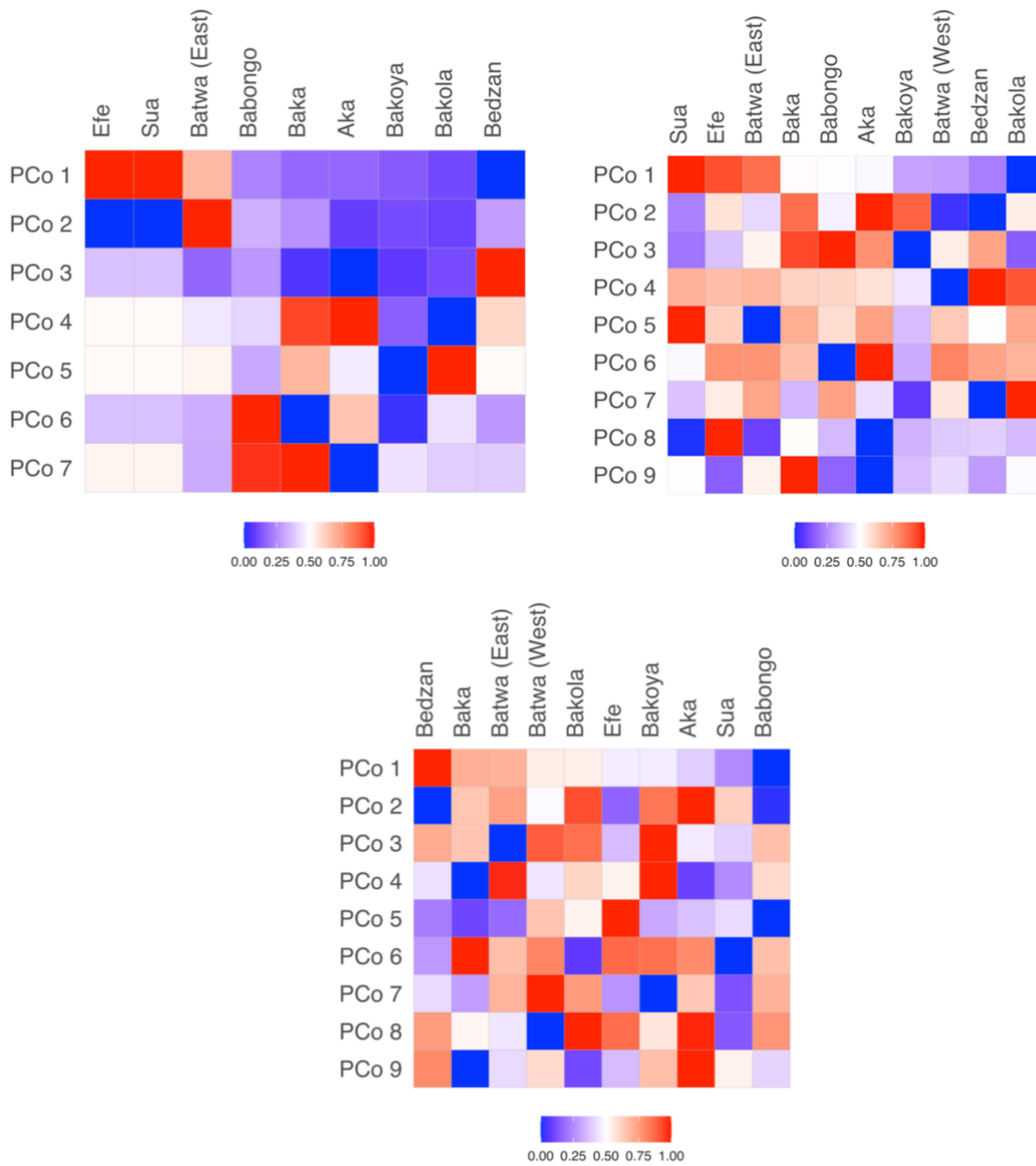


Figure 18. Contribution to PCoA heatmaps in genes (top left), music (top right) and subsistence toolkits (bottom)

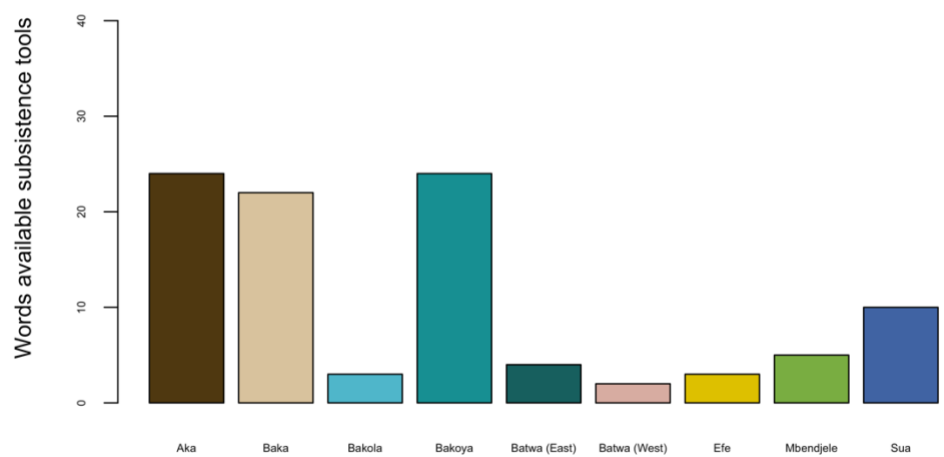
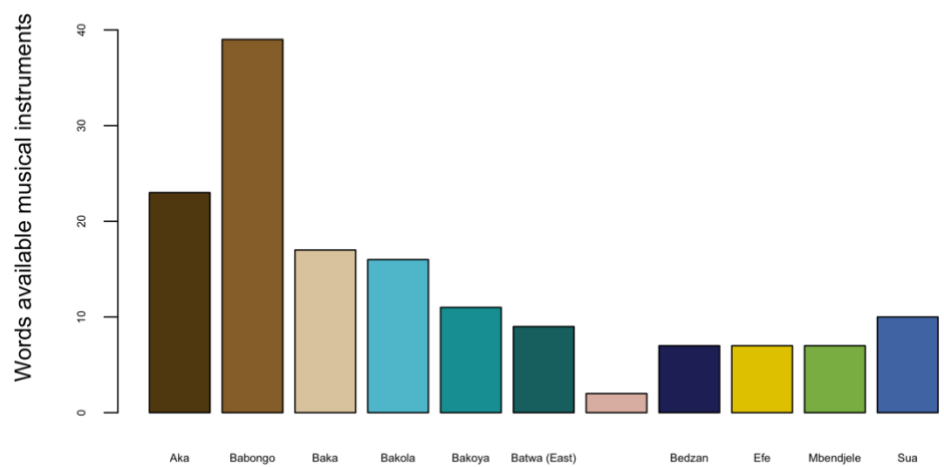


Figure 19. Number of different words compiled per population to designate musical instruments (top) and subsistence tools (bottom)

Biomes

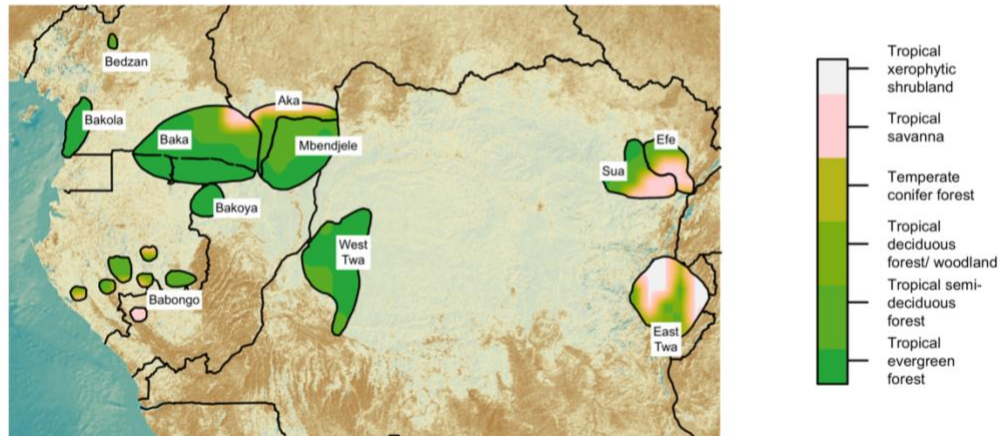
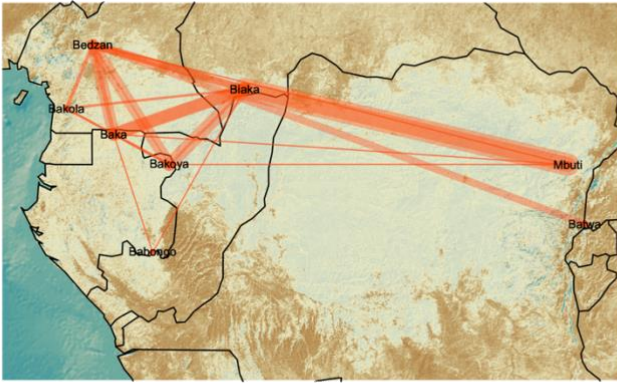
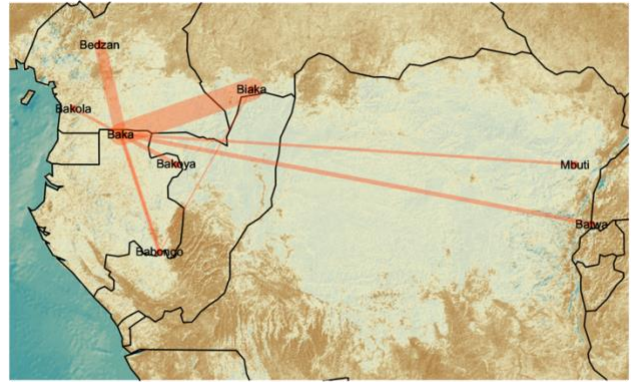


Figure 20. Biomes in the territories of the cultural groups included in our analyses obtained from Beyer *et al.* (2020)

1-5cM



5-10cM



>10cM

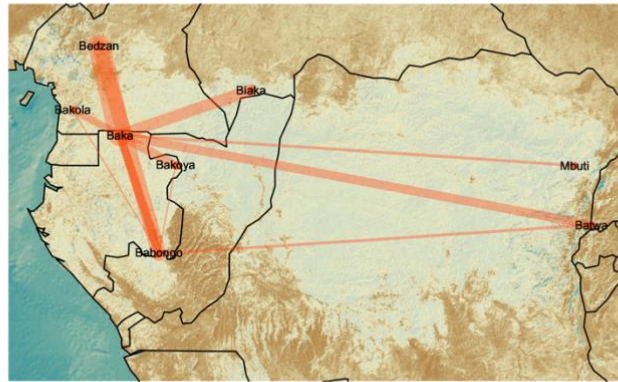


Figure 21. Recent genetic connectivity between CAHG populations. Network visualizations of the average number of IBD segments shared per crosspopulation individual pairs using only CAHG genetic segments with identified IBD blocks in the range of: (A) 1 to 5 cM (2,500 to 1,500 y ago), (B) 5 to 10 cM (1,500 to 500 y ago), (C) and over 10cM (500 to 0 y ago). Thicker lines indicate greater gene flow as identified by a higher probability of sharing IBD blocks of the specified length.

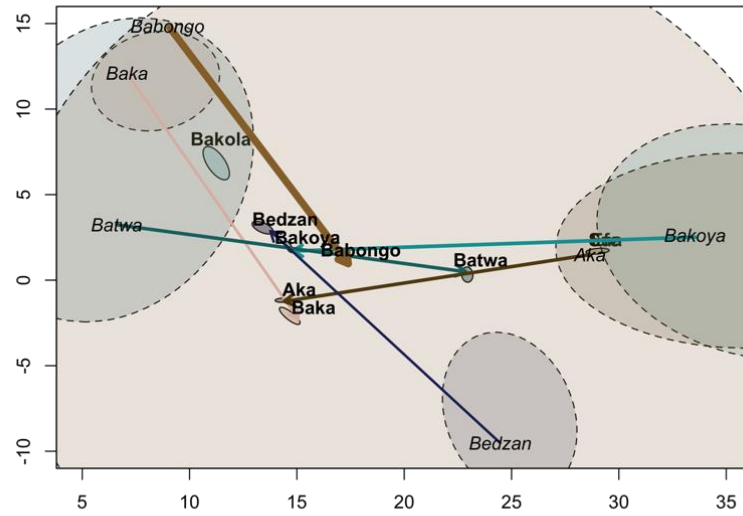


Figure 22. Estimated recent gene flow between CAHG by SpaceMix. Bold letters indicate geo-genetic coordinates of populations in our sample, *Italic letters* indicate sources of admixture, and arrows indicate direction of admixture. Ellipses bound by solid lines indicate 95% confidence interval around the estimated geo-genetic location of each population and ellipses bound by dashed lines 95% confidence intervals around the hypothesised admixture source. Width of arrows is proportional to estimated proportion of ancestry of a population deriving from admixture source location (origin of the arrows).

Table 1. Area, and location of each group after bahichet 2006;2012 and estimated population size of each of the groups by Bahuchet 2006; Hewlett and Fancher, 2013 (Batwa west), and Lewis, 2000 (for the Batwa of the great lakes). For Babongo, we merged his estimate for central and southern Babongo.

| Culture | Longitude | Latitude | Area | Estimated census pop size | Country |
|--------------|-----------|----------|------|---------------------------|------------------------|
| Aka | 17.22 | 2.43 | 6.09 | 30,000-50,000 | CAR, Congo |
| Babongo | 11.98 | -2.00 | 1.84 | 9,500 | Gabon |
| Baka | 14.14 | 2.35 | 8.01 | 30,000-40,000 | Cameroon, Gabon, Congo |
| Bakola | 10.16 | 3.01 | 1.12 | 4,000 | Cameroon |
| Bakoya | 14.49 | 0.70 | 0.88 | 2,600 | Gabon, Congo |
| Batwa (East) | 29.35 | -2.38 | 4.45 | 70,000-87,000 | DRC, Uganda, Rwanda |
| Batwa (West) | 18.53 | -1.33 | 4.89 | 14,000 | DRC |
| Bedzan | 11.36 | 5.87 | 0.11 | 400 | Cameroon |
| Efe | 29.33 | 1.81 | 1.73 | 10,000 | DRC |
| Sua | 28.38 | 1.62 | 2.44 | 26,000 | DRC |

Table 2. Populations included in our genetic analyses. Asterisks indicate populations were included in the reduced dataset we created for running ADMIXTURE as well as in our IBDNe analyses

| Population | N | Meta group | ReducedData | IBDNe |
|------------|-----|------------|-------------|-------|
| Boni | 21 | HG | | * |
| Hadza | 26 | HG | | * |
| Ogiek | 17 | HG | | * |
| Sandawe | 29 | HG | | * |
| Wata | 4 | HG | | |
| Babongo | 50 | West CAHG | * | * |
| Baka | 144 | West CAHG | * | * |
| Bakola | 29 | West CAHG | * | * |
| Bakoya | 20 | West CAHG | * | * |
| Bedzan | 24 | West CAHG | * | * |
| Aka | 17 | West CAHG | * | * |
| Batwa | 35 | East CAHG | * | * |
| Mbuti | 13 | East CAHG | * | * |
| Aari | 9 | Non_HG | | |
| Ahizi | 17 | Non_HG | | |
| Akele | 39 | Non_HG | | * |
| Badwee | 38 | Non_HG | | * |
| Bakiga | 54 | Non_HG | * | * |
| Bakota | 47 | Non_HG | | |

| | | | |
|-----------|----|--------|---|
| Baniamer | 10 | Non_HG | |
| Bapunu | 47 | Non_HG | |
| Bariba | 20 | Non_HG | * |
| Bateke | 43 | Non_HG | |
| Bekwil | 5 | Non_HG | |
| Benga | 40 | Non_HG | * |
| Borana | 18 | Non_HG | |
| Bulala | 12 | Non_HG | |
| Burji | 15 | Non_HG | |
| Datog | 18 | Non_HG | |
| Dinka | 12 | Non_HG | |
| Duma | 39 | Non_HG | * |
| Elmolo | 13 | Non_HG | |
| Eshira | 40 | Non_HG | * |
| Eviya | 20 | Non_HG | * |
| Fang | 67 | Non_HG | * |
| Fon | 12 | Non_HG | * |
| Fulani | 24 | Non_HG | |
| Gabra | 9 | Non_HG | |
| Galoa | 48 | Non_HG | * |
| Gurreh | 7 | Non_HG | |
| Hadandawa | 15 | Non_HG | |
| Hamer | 10 | Non_HG | |
| Ilchamus | 19 | Non_HG | |
| Iraqw | 23 | Non_HG | |
| Iyassa | 20 | Non_HG | |
| Kikuyu | 14 | Non_HG | |
| Kimbundu | 18 | Non_HG | |
| Kongo | 11 | Non_HG | |
| Lemande | 19 | Non_HG | |
| Luo | 32 | Non_HG | |
| Mada | 19 | Non_HG | |
| Makina | 40 | Non_HG | * |
| Mbororo | 16 | Non_HG | |
| Ndumu | 38 | Non_HG | * |
| Ngumba | 20 | Non_HG | |
| Nzebi | 61 | Non_HG | * |
| Nzime | 42 | Non_HG | * |
| Obamba | 45 | Non_HG | * |
| Okande | 6 | Non_HG | |
| Orma | 14 | Non_HG | |
| Orungu | 20 | Non_HG | |

| | | | |
|-----------|----|--------|---|
| Ovimbundu | 16 | Non_HG | |
| Pare | 14 | Non_HG | |
| Pokot | 5 | Non_HG | |
| Rangi | 14 | Non_HG | |
| Rendille | 14 | Non_HG | |
| Sabue | 14 | Non_HG | |
| Sanye | 14 | Non_HG | |
| Sengwer | 16 | Non_HG | |
| Shake | 46 | Non_HG | * |
| Taita | 9 | Non_HG | |
| Taveta | 2 | Non_HG | |
| Tikar | 19 | Non_HG | |
| Tsogo | 62 | Non_HG | |
| Tugen | 15 | Non_HG | |
| Yaaku | 16 | Non_HG | |
| Yacuba | 17 | Non_HG | |
| Yaounde | 40 | Non_HG | |
| Yoruba | 32 | Non_HG | * |

Table 3. Mean proportion of ancestry components from out ADMIXTURE run at $K=3$. CAHG_1 and CAHG_2 refer to ancestry components associated with Western and Eastern CAHG respectively. AGR refer to ancestry components associated with African agriculturalist populations

| Population | CAHG_1 | CAHG_2 | AGR |
|------------|--------|--------|----------|
| Babongo | 0.378 | 0.100 | 0.523 |
| Baka | 0.851 | 0.012 | 0.137 |
| Bakola | 0.663 | 0.109 | 0.228 |
| Bakoya | 0.656 | 0.110 | 0.234 |
| Batwa | 0.014 | 0.768 | 0.219 |
| Bedzan | 0.522 | 0.078 | 0.400 |
| Aka | 0.763 | 0.080 | 0.157 |
| Efe & Sua | 0.225 | 0.775 | 0.000001 |

Table 4. Average heterozygosity of populations in our sample using unmasked dataset

| Population | Heterozygosity |
|--------------|----------------|
| Babongo | 0.326 |
| Baka | 0.319 |
| Bakola | 0.318 |
| Bakoya | 0.318 |
| Batwa (East) | 0.315 |
| Bedzan | 0.323 |
| Aka | 0.317 |
| Efe & Sua | 0.291 |

Table 5. Mantel tests predicting structure of genetic diversity (F_{ST}) using CAHG ancestry components. P values are adjusted with Benjamini Hochberg procedure.

| | Genetic diversity (F_{ST}) | |
|---------------------------|--------------------------------|--------------|
| | Statistic | Adj p |
| Geography | 0.818 | 0.001 |
| Ecology | 0.434 | 0.022 |
| Geography (cont. ecology) | 0.746 | 0.001 |
| Ecology (cont. geography) | 0.201 | 0.182 |

Table 6. Number of simulated SNPs for sequence data generated to validate IBDNe results before and after minor allele frequency filtering.

| Model | Sample size | SNPs | SNPs MAF=0.05 | SNPs MAF=0.1 |
|-----------|-------------|----------|---------------|--------------|
| collapse | 15 | 27765599 | 20664486 | 16895493 |
| collapse | 20 | 26680735 | 22137250 | 16543917 |
| collapse | 50 | 35811285 | 21580546 | 15931823 |
| collapse | 150 | 42370877 | 21056808 | 15674009 |
| constant | 15 | 6279121 | 4635268 | 3788050 |
| constant | 20 | 6187033 | 5112965 | 3693104 |
| constant | 50 | 8195115 | 4845000 | 3573145 |
| constant | 150 | 9943843 | 4728448 | 3514107 |
| expansion | 15 | 7102193 | 5149201 | 4207589 |
| expansion | 20 | 7666131 | 5681552 | 4122339 |
| expansion | 50 | 9553205 | 5379826 | 3969557 |
| expansion | 150 | 12376707 | 5243012 | 3896819 |

Table 7. Estimated admixture proportion from hypothesised admixture source location in Spacemix (taken from MCMC run with highest posterior probability)

| Population | Proportion of genome from inferred admixture source |
|--------------|---|
| Baka | 0.110 |
| Bakoya | 0.054 |
| Bakola | 0.002 |
| Efe & Sua | 0.002 |
| Batwa (East) | 0.063 |
| Aka | 0.061 |
| Bedzan | 0.128 |
| Babongo | 0.010 |

Table 8. Multiple matrix regressions assessing the relationship between geographical, genetic, ecological and cultural distance with masked dataset (CAHG genomic segments).

| | Musical instruments | | Subsistence tools | |
|---------------------------|---------------------|----------------|-------------------|----------------|
| | Coefficient | <i>p</i> value | Coefficient | <i>p</i> value |
| Genes (cont. Geography) | 0.185 | 0.191 | 0.244 | 0.164 |
| Genes (cont. ecology) | 0.414 | 0.018 | 0.142 | 0.274 |
| Geography (cont. Genes) | 0.269 | 0.088 | -0.031 | 0.578 |
| Geography (cont. Ecology) | 0.395 | 0.008 | 0.0457 | 0.373 |
| Ecology (cont. Geography) | -0.029 | 0.559 | 0.496 | 0.015 |
| Ecology (cont. Genes) | -0.033 | 0.541 | 0.309 | 0.119 |

Table 9. Total number of shared IBD segments >1cM between individuals belonging to CAHG populations (only within CAHG genomic components)

| | Babongo | Baka | Bakola | Bakoya | Batwa | Bedzan | Aka | Efe & Sua |
|-----------|---------|-------|--------|--------|-------|--------|-------|-----------|
| Babongo | 1385 | 2790 | 394 | 330 | 378 | 639 | 527 | 68 |
| Baka | 2790 | 71819 | 4549 | 4118 | 1714 | 12155 | 28922 | 339 |
| Bakola | 394 | 4549 | 981 | 835 | 390 | 1718 | 1325 | 32 |
| Bakoya | 330 | 4118 | 835 | 882 | 284 | 1623 | 1386 | 31 |
| Batwa | 378 | 1714 | 390 | 284 | 4245 | 393 | 213 | 100 |
| Bedzan | 639 | 12155 | 1718 | 1623 | 393 | 9904 | 4735 | 127 |
| Aka | 527 | 28922 | 1325 | 1386 | 213 | 4735 | 7358 | 77 |
| Efe & Sua | 68 | 339 | 32 | 31 | 100 | 127 | 77 | 3015 |

Table 10. Total length of IBD segments >1cM among shared all individuals from each CAHG population pair (only within CAHG genomic components)

| | Babongo | Baka | Bakola | Bakoya | Batwa | Bedzan | Aka | Efe & Sua |
|-----------|-----------|------------|-----------|-----------|-----------|-----------|-----------|-----------|
| Babongo | 16893.098 | 12601.295 | 1968.582 | 1650.36 | 2423.201 | 1664.461 | 1114.199 | 128.321 |
| Baka | 12601.295 | 254052.515 | 15724.828 | 13071.769 | 8204.875 | 29727.654 | 78632.294 | 683.507 |
| Bakola | 1968.582 | 15724.828 | 4172.076 | 3090.363 | 2955.117 | 4285.595 | 2980.787 | 66.201 |
| Bakoya | 1650.36 | 13071.769 | 3090.363 | 3590.363 | 2328.025 | 3976.952 | 3209.948 | 57.697 |
| Batwa | 2423.201 | 8204.875 | 2955.117 | 2328.025 | 20928.371 | 1379.269 | 443.288 | 214.79 |
| Bedzan | 1664.461 | 29727.654 | 4285.595 | 3976.952 | 1379.269 | 64284.796 | 11427.79 | 247.443 |
| Aka | 1114.199 | 78632.294 | 2980.787 | 3209.948 | 443.288 | 11427.79 | 31292.914 | 149.019 |
| Efe & Sua | 128.321 | 683.507 | 66.201 | 57.697 | 214.79 | 247.443 | 149.019 | 9804.437 |

Table 11. Linguistic classification of the languages spoken by the CAHG populations included in this study from Bahuchet 2006;2012 (Following classification by Greenberg, 1966; Ruhen, 1991 and Guthrie, 1967-71).

| CAHG group | Phylum | Stock | Family | Group | Sub-group |
|--------------|-------------------|-----------------|---------------|--------------------|---------------|
| Baka | Nigel-Kordofanian | Niger-Congo | North Central | Adamawa-Ubangian | Gbanzili-Sere |
| Bedzan | Nigel-Kordofanian | Niger-Congo | South Central | Bantoid, Non-Bantu | Tikar |
| Bakola | Nigel-Kordofanian | Niger-Congo | South Central | Northwest Bantu | A80 |
| Bakoya | Nigel-Kordofanian | Niger-Congo | South Central | Northwest Bantu | B20 |
| Babongo | Nigel-Kordofanian | Niger-Congo | South Central | Northwest Bantu | B30 |
| Babongo | Nigel-Kordofanian | Niger-Congo | South Central | Northwest Bantu | B60 |
| Babongo | Nigel-Kordofanian | Niger-Congo | South Central | Northwest Bantu | B70 |
| Mbendjele | Nigel-Kordofanian | Niger-Congo | South Central | Northwest Bantu | C10 |
| Aka | Nigel-Kordofanian | Niger-Congo | South Central | Northwest Bantu | C10 |
| Batwa (West) | Nigel-Kordofanian | Niger-Congo | South Central | Northwest Bantu | C60 |
| Batwa (East) | Nigel-Kordofanian | Niger-Congo | South Central | Central Bantu | J11 |
| Batwa (East) | Nigel-Kordofanian | Niger-Congo | South Central | Central Bantu | J60 |
| Sua | Nigel-Kordofanian | Niger-Congo | South Central | Central Bantu | D30 |
| Efe | Nilo-Saharan | Central-Sudanic | East Central | Mangbetu-Efe | - |

Table 12. Shared lexical items between CAHG groups

| Word | Meaning | Populations sharing word and meaning |
|----------------------------|---------------------|---|
| Musical instruments | | |
| ndumu | drum (conical) | Aka, Baka, Bakoya, Mbendjele |
| ngom | drum (cylindrical) | Bakoya, Bakola, Batwa (West), Babongo |
| hindehu | flute | Aka, Mbendjele |
| ngombi | harp | Babongo, Baka, Aka |
| bogongo | zither harp | Aka, Baka |
| ligbebe | rattle | Aka, Baka |
| soko | rattle | Aka, Babongo |
| baka | struck beam | Babongo, Baka |
| mobio | flute | Aka, Baka |
| ruma | flute | Efe, Sua |
| sanze | lamellaphone | Aka, Babongo |
| mokinda | single-skinned drum | Aka, Baka |
| kembe | lamellaphone | Baka, Batwa (East), Efe, Sua |
| Subsistence tools | | |
| apekulo | cable snare | Baka, Bakoya |
| api | arrow | Sua, Efe |
| sàwàla | leather sac | Baka, Aka |
| mokobe | honey container | Baka, Aka |